



UNIVERSITY OF
BIRMINGHAM

Bloodwise
Beating blood cancer since 1960

UKALL 2011 Trial

United Kingdom National Randomised Trial For Children and Young Adults with Acute Lymphoblastic Leukaemia and Lymphoma 2011

Version 8.0
Date 19th January 2022

Sponsor: University of Birmingham
Sponsor Protocol Number: RG_09-072

CAS CODE: HM3009
Eudract Number: 2010-020924-22
ISRCTN Reference Number: ISRCTN64515327

Chief Investigator:

Professor Ajay Vora
Consultant Paediatric Haematologist
Paediatric Haematology
Great Ormond Street Hospital
London
WC1N 3JH

TABLE OF CONTENTS

Introductory pages/Contacts	7
Chief Investigator Signature Page.....	14
Amendments.....	15
Trial Synopsis.....	23
Title	23
Trial Design.....	23
Aims	23
Objectives	23
Outcome Measures.....	23
Patient Population	24
Sample Size	24
Main Inclusion and Exclusion Criteria.....	25
Trial Duration.....	25
Treatment Summary	26
Trial Schema.....	29
Schedule of Events for ALL patients.....	30
Schedule of Events for LBL patients.....	32
Abbreviations.....	33
1. Background and Rationale.....	35
1.1 Background.....	35
1.2 Trial Rationale.....	37
1.2.1 Treatment stratification	38
1.2.2 Rationale for Use of Dexamethasone.....	41
1.2.3 Rationale for Dexamethasone Randomisation.....	42
1.2.4 Rationale for use of split dexamethasone in patients aged 10 and over.....	43
1.2.5 Rationale for high dose methotrexate randomisation.....	44
1.2.6 Rationale for Pulses randomisation.....	45
1.2.7 High Risk Cytogenetics in B-cell precursor ALL (See also appendix 25 for information on testing for ABL-class fusion abnormalities).....	47
1.2.8 Definition of good risk cytogenetic abnormalities in B-cell precursor ALL.....	48
2. Aims, Objectives and Outcome Measures	49
2.1 Aims and Objectives	49
2.1.1 Aims.....	49
2.1.2 Objectives	49
2.2 Outcome Measures	50
2.2.1 Primary outcome measures.....	50
2.2.2 Secondary outcome measures.....	50
3. Trial Design.....	51
3.1 Changes to treatment regimens compared to UKALL 2003	51
3.2 Stratification of treatment in UKALL 2011	51
3.2.1 Acute Lymphoblastic Leukaemia.....	51
3.2.2 Lymphoblastic Lymphoma.....	52
3.3 Allocation of induction therapy.....	53
3.3.1 Modification of Induction for High Risk Cytogenetics	53
3.3.2 Induction for Down Syndrome patients (ALL and LBL)	54
3.4 Allocation of post-induction therapy in patients with ALL	54

3.4.1	Obtaining a Day 29 MRD result.....	54
3.4.2	Allocation of post-induction therapy (Day 29 MRD Result)	54
3.4.3	Obtaining a post consolidation MRD result (MRD Risk group only).....	55
3.4.4	Allocation of post consolidation therapy (MRD Risk group only)	56
3.5	Allocation of post-induction therapy in LBL patients.....	56
3.6	Methotrexate and Pulses Randomisation (R2).....	57
3.7	Special Categories	58
3.7.1	CNS Disease at Presentation (CNS3):.....	58
3.7.2	Traumatic or CNS2 LP at diagnosis	58
3.7.3	Testicular Disease at Diagnosis	58
3.7.4	High risk cytogenetics.....	59
3.7.5	Induction Failure: defined as M3 marrow and/or molecular MRD >5%. In the absence of molecular MRD, M2 marrow confirmed on central review by CI or co-investigators.	59
3.8	Duration of treatment.....	60
3.9	Frequency and duration of follow-up	60
4.	Eligibility	61
4.1	Inclusion Criteria.....	61
4.1.1	Trial entry.....	61
4.1.2	Exclusion criteria for R2 (methotrexate and pulses randomisation).....	61
5.	Screening and Consent	62
5.1	Screening.....	62
5.1.1	Collection of diagnostic samples from LBL patients at diagnosis	63
5.2	Informed Consent	63
5.2.1	Stages of consent	64
6.	Trial Entry.....	65
6.1	Procedure for online patient registration and randomisation.....	65
6.2	Emergency registrations and randomisations	66
7.	Treatment Details	67
7.1	Risks of intrathecal therapy	67
7.2	General management of all treatment regimens	67
7.2.1	Registration.....	67
7.2.2	Randomisation 2 (R2).....	67
	End of treatment.....	68
7.3	Trial Treatment	69
7.3.1	Non-Investigational Medicinal Products (NIMPs)	70
7.4	Determining weight for dosage calculations.....	71
7.4.1	Children (patients aged under 18 years)	71
7.4.2	Adults (patients aged 18 and above).....	72
7.5	Regimen A Treatment Schedule	73
7.5.1	Summary of Regimen A Treatment.....	73
	Figure 6: Allocation of induction treatment in Regimen A.....	Error! Bookmark not defined.
7.5.2	Regimen A: Induction	74
7.5.3	Regimen A: Post-induction therapy	78
7.5.4	Regimen A: Consolidation	79
7.5.5	Regimen A: Standard Interim Maintenance	80
7.5.6	Regimen A: High Dose Methotrexate (Protocol M)	81
7.5.7	Regimen A: Delayed Intensification	82

7.5.8	Regimen A: Maintenance	84
7.6	Regimen B Treatment Schedule	89
7.6.1	Summary of Regimen B Treatment	89
7.6.2	Regimen B: Induction	90
7.6.3	Regimen B: Post-induction therapy	93
7.6.4	Regimen B: Standard BFM Consolidation	94
7.6.5	Regimen B: Standard Interim Maintenance	95
7.6.6	Regimen B: High Dose Methotrexate (Protocol M)	96
7.6.7	Regimen B: Delayed Intensification	98
7.6.8	Regimen B: Maintenance	100
	Regimen C Treatment Schedule	107
7.6.9	Summary of Regimen C Treatment	107
7.6.10	Regimen C: Induction	108
7.6.11	Regimen C: Post-induction therapy	111
7.6.12	Regimen C: Augmented BFM Consolidation	113
7.6.13	Regimen C: Capizzi Interim Maintenance	115
7.6.14	Regimen C: High dose methotrexate + asparaginase (Protocol M-A)	117
7.6.15	Regimen C: Delayed Intensification	119
7.6.16	Regimen C: Maintenance	121
7.7	Assessments	129
7.7.1	Routine Clinical and Laboratory Assessments during treatment	129
7.8	Sample Collection	129
7.8.1	Diagnosis	129
7.8.2	During induction	131
7.8.3	During Consolidation	131
7.8.4	During Maintenance	132
7.9	MRD sample requirements	132
7.10	Quality of Life questionnaires	132
7.11	Dose Modifications	133
7.11.1	Dexamethasone	133
7.11.2	Mercaptopurine (6-MP) and methotrexate (MTX)	133
7.11.3	Co-trimoxazole (trimethoprim and sulphamethoxazole)	135
7.12	Dose modifications for toxicity	136
7.13	Supportive treatment	142
7.13.1	Tumour Lysis	142
7.13.2	Antimicrobials	142
7.13.3	Antiemetics	142
7.13.4	Mouthcare	142
7.13.5	G-CSF	143
7.14	Patient Follow Up	143
7.15	Patient Withdrawal	143
7.15.1	Withdrawal from UKALL 2011 trial treatment	143
7.15.2	Withdrawal of consent to data collection	143
7.15.3	Lost to follow-up	143
7.16	Relapse	143
8.	Adverse Event Reporting	144
8.1	Reporting Requirements	144

8.1.1	Adverse Events.....	144
8.1.2	Serious Adverse Events	144
8.1.3	Reporting period	147
8.2	Reporting Procedure	147
8.2.1	Site.....	147
8.2.2	Trials Office.....	148
8.2.3	Reporting to the Competent Authority and main Research Ethics Committee	148
8.2.4	Investigators	149
8.2.5	Data Monitoring Committee.....	149
9.	Data Handling and Record Keeping	149
9.1	Data Collection	149
9.1.1	Registered patients data collection.....	149
9.1.2	Data collection for registered patients who decline R2	149
9.1.3	Ad hoc forms.....	149
9.2	Archiving	150
10.	Quality Management	150
10.1	Site Set-up and Initiation	150
10.2	On-site Monitoring	150
10.3	Central Monitoring	151
10.4	Audit and Inspection	151
10.5	Notification of Serious Breaches	151
11.	End of Trial Definition	151
12.	Statistical Considerations	151
12.1	Randomisation.....	151
12.2	Arrangements for protecting against bias.....	152
12.3	Definition of Outcome Measures	152
12.3.1	Primary outcome measures.....	152
12.3.2	Secondary outcome measures.....	152
12.4	Analysis of Outcome Measures.....	153
12.5	Planned Sub Group Analyses	153
12.6	Planned Interim Analysis	153
12.7	Stopping Guidelines	154
12.8	Planned Final Analyses	154
12.9	Power Calculations.....	154
13.	Trial Organisational Structure	155
13.1	Sponsor	155
13.2	Coordinating Centre	155
13.3	Relationship of trial committees.....	155
13.4	Trial Management Group.....	156
13.5	Trial Steering Committee.....	156
13.6	Data Monitoring Committee.....	156
13.7	Finance	156
14.	Ethical Considerations	156
15.	Confidentiality and Data Protection	157
16.	Insurance and Indemnity.....	157
17.	Publication Policy	157
18.	Reference List.....	158

Appendix 1 - WMA Declaration of Helsinki	161
Appendix 2 - Definition of Adverse Events.....	164
Appendix 3 - Common Toxicity Criteria Gratings	166
Appendix 4 – Measurement of MRD in UKALL 2011.....	167
Appendix 5 - Treatment modifications for Down Syndrome Patients	171
Appendix 6 - Drug Interactions With Cytotoxic Agents	173
Appendix 7 - Administration of High Dose Methotrexate.....	177
Appendix 8 - Management of Encephalopathy related to intrathecal methotrexate	181
Appendix 9 - Guidelines for the investigation and management of thrombo-embolic events..	182
Appendix 10 - Testicular radiotherapy guidelines	185
Appendix 11– Health economics	186
Appendix 12 – Asparaginase study- CLOSED	187
Appendix 13 – Quality of life study.....	191
Appendix 14 – Flow MRD Study –Closed	195
Appendix 15 – Dexamethasone pharmacokinetic study - CLOSED.....	198
Appendix 16 – Investigational Medicinal Product (IMP) Supplies.....	201
Appendix 17 – Osteonecrosis Recommendations.....	202
Appendix 18 – Diagnosis And Management Of Pancreatitis	203
Appendix 19 – Use Of Crisantaspase (Erwinia L-asparaginase) In Patients With Systemic Reactions To Pegaspargase (Oncaspar)	204
Appendix 20 – Genomics Sub-study.....	206
Appendix 21 –TPMT and 6mp	208
Appendix 22 – T-Cell NHL Emergencies	210
Appendix 23 – Diagnostic Flow Cytometry in UKALL 2011	212
Appendix 24 – Vincristine pharmacokinetic and pharmacogenomic study - CLOSED	213
Appendix 25 – Screening for targetable tyrosine kinase alterations in UKALL2011 patients ..	217
Appendix 26 – Chemotherapy –Associated Neurotoxicity Study	220
Appendix 27 TRECA (TRials Engagement in Children and Adolescents) Study – Open at Selected UKALL 2011 Sites	227

INTRODUCTORY PAGES/CONTACTS

Protocol Title	United Kingdom National Randomised Trial For Children and Young Adults with Acute Lymphoblastic Leukaemia and Lymphoma 2011
Protocol Short Name	UKALL 2011
Protocol Version	8.0, 19 th January 2022
EudraCT number	2010-020924-22
Sponsor Protocol Number	RG_09-072
CAS Number	HM3009
ISRCTN Reference Number	ISRCTN64515327
Sponsor	Cancer Research UK Clinical Trials Unit (CRCTU) University of Birmingham Edgbaston, Birmingham, B15 2TT, UK
Funded by	Bloodwise

Chief Investigator:

Professor Ajay Vora
 Consultant Paediatric Haematologist
 Paediatric Haematology
 Great Ormond Street Hospital
 London
 WC1N 3JH
 ☎ 02077626085
 ✉ ajay.vora@gosh.nhs.uk
 📠 02078138410

Co-investigators for patients aged <16 years:

Dr John Moppett
Consultant Paediatric Haematologist
Bristol Royal Hospital for Children
Upper Maudlin Street Bristol,
BS2 8BJ
☎ 0117 342 8752
✉ john.moppett@UHBristol.nhs.uk
📠 0117 342 8628

Dr Sujith Samarasinghe
Consultant Paediatric Haematologist
Level 2 Camelia Botnar Laboratory
Great Ormond Street Hospital
Great Ormond Street London
WC1N 3JH
☎ 0207 829 8190
✉ sujith.samarasinghe@gosh.nhs.uk
📠 0207 813 8410

Dr Sarah Lawson
Consultant Paediatric Haematologist
Birmingham Children's Hospital
Steelhouse Lane
Birmingham
West Midlands
B4 6NH.
☎ 0121 333 9847
✉ sarah.lawson@bch.nhs.uk
📠 0121 333 9841

Associate Co-investigators for patients aged <16 years

Dr Katharine Patrick
Consultant Paediatric Haematologist
Dept. of Haematology
Sheffield Children's Hospital
Western Bank
Sheffield
S10 2TH
☎ 0114 271 7477
✉ katharine.patrick@sch.nhs.uk
📠 0114 276 2289

Emeritus Investigator

Dr Nick Goulden
Contact via Professor Vora

Co-investigators for patients aged 16 – 24 years:

Dr Clare Rowntree
Consultant Haematologist
University Hospital of Wales
Cardiff CF14 4XN
☎ 029 20744327

✉ clare.rowntree@wales.nhs.uk

☎ 02920 746427

Dr Rachael Hough
Consultant Haematologist
University College Hospital,
1st Floor West Wing, 6th Floor Central
250 Euston Road,
London.
NW1 2PQ

☎ 020 7380 9952

✉ rachael.hough@uclh.nhs.uk

☎ 020 7380 9064

Co-investigators for lymphoblastic lymphoma (LBL):

Dr Robert Wynn
Consultant Paediatric Haematologist
Royal Manchester Children's Hospital
Oxford Road
Manchester
M13 9WL

☎ 0161 701 8417

✉ robert.wynn@cmft.nhs.uk

☎ 0161 701 8410

Trial Statistician:

Amy Kirkwood
Cancer Research UK & UCL Cancer Trials Centre
University College London
90 Tottenham Court Road (5th Floor)
London W1T 4TJ

☎ 0207 679 9231

✉ a.kirkwood@ucl.ac.uk

☎ 0207 679 9861

Pharmacy Advice:

Please e-mail any pharmacy-related queries to the trial mailbox:

☎ 0121 414 7626

✉ ukall2011@trials.bham.ac.uk

Molecular MRD Coordinators:

Clinical

Dr John Moppett
Consultant Paediatric Haematologist
Bristol Royal Hospital for Children
Upper Maudlin Street Bristol,
BS2 8BJ

☎ 0117 342 8752

✉ john.moppett@UHBristol.nhs.uk

☎ 0117 342 8628

Molecular MRD Coordinators contd:**Laboratory**

Dr Jeremy Hancock
 Bristol Genetics Laboratory
 Pathology Sciences
 Southmead Hospital
 Westbury-on-Trym
 Bristol BS10 5NB
 ☎ 0117 323 6262
 ✉ jeremy.hancock@nbt.nhs.uk
 📠 0117 323 5572

Cytogenetics Coordinating Centre

Leukaemia Research Cytogenetics Group (LRCG)
 Northern Institute for Cancer Research
 Newcastle University
 Level 5, Sir James Spence Institute,
 Royal Victoria Infirmary
 Queen Victoria Road
 Newcastle upon Tyne
 NE1 4LP
 ☎ 0191 282 1320 (Professor Christine Harrison) or
 0191 282 1323 (Professor Anthony Moorman)
 ✉ christine.harrison@newcastle.ac.uk,
 ✉ anthony.moorman@newcastle.ac.uk
 lrcg-clinical@newcastle.ac.uk
 📠 0191 282 1326

Quality of Life Study:

Dr Meriel Jenney
 Children's Hospital for Wales
 Heath Park
 Cardiff
 Wales
 CF14 4XW.
 ☎ 0292 074 2107
 ✉ meriel.jenney@wales.nhs.uk
 📠 0292 074 5395

Vincristine Study

Dr. Sujith Samarasinghe
 Consultant Paediatric Haematologist
 Level 2 Camelia Botnar Laboratory
 Great Ormond Street Hospital
 Great Ormond Street London
 WC1N 3JH
 ☎ 0207 829 7918
 ✉ Sujith.samarasinghe@gosh.nhs.uk

Neurotoxicity Study

Dr Christina Halsey
Clinical Senior Lecturer & Honorary NHS Consultant
Paediatric Haematologist
Wolfson Wohl Cancer Research Centre,
University of Glasgow
Glasgow
G61 1QH UK
☎ 0141 330 8135
📠 0141 942 6521
✉ chris.halsey@glasgow.ac.uk

**TRECA (TRials Engagement in
Children and Adolescents) Study**

Dr Peter Knapp
TRECA Chief Investigator
Department of Health Sciences
Seebohm Rowntree Building
University of York
Heslington
York
YO10 5DD
☎ 01904 321675
✉ peter.knapp@york.ac.uk

Dr Jackie Martin-Kerry
TRECA Study Manager
Department of Health Sciences
Seebohm Rowntree Building
Heslington
York
YO10 5DD
☎ 01904 321826
✉ jackie.martin-kerry@york.ac.uk

Asparaginase Study:

Professor Vaskar Saha
Institute of Cancer Sciences
The University of Manchester
Manchester Academic Health Science Centre
Christie Hospital
Wilmslow Rd
Manchester
M20 4BX
☎ 0161 446 3023
✉ pao@manchester.ac.uk
📠 0161 446 3092

Dexamethasone Study:

Dr Gareth Veal
Northern Institute for Cancer Research
University of Newcastle
Medical School
Newcastle upon Tyne
NE2 4HH
☎ 0191 246 4332
✉ g.j.veal@newcastle.ac.uk
📠 0191 222 3452

Flow MRD Study:

Dr Sarah Lawson
Consultant Paediatric Haematologist
Birmingham Children's Hospital
Steelhouse Lane
Birmingham
West Midlands
B4 6NH.
☎ 0121 333 9847
✉ sarah.lawson@bch.nhs.uk
📠 0121 333 9841

Trial Coordinator & Trials Office:

Jon Mee
Children's Cancer Trials Team
Cancer Research UK Clinical Trials Unit (CRCTU)
University of Birmingham
School of Cancer Sciences
Vincent Drive
Edgbaston
Birmingham B15 2TT
☎ 0121 414 7626
✉ ukall2011@trials.bham.ac.uk
☎ 0121 414 9520

Randomisation Service:

Provided by the CRCTU at the University of Birmingham.

Randomisation should be performed by sites online at
<https://www.cancertrials.bham.ac.uk/UKALL2011Live>

In case of any problems with online randomisation, randomisation can be performed over the phone by the CRCTU on:

☎: 0121 414 7626 or 0121 415 1068

SAE reporting:

SAEs should be faxed to the UKALL 2011 Trials Office, CRCTU, University of Birmingham.

☎ 0121 414 9520 or 0121 414 3700

If reporting by fax is not possible, please send to:

Reg@trials.bham.ac.uk

Include "UKALL2011 SAE" in the subject line

CHIEF INVESTIGATOR SIGNATURE PAGE

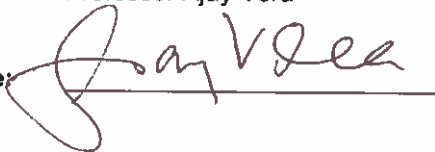
UKALL 2011 Trial Protocol Version 8.0, 19th January 2022

This protocol has been approved by:

Name: Professor Ajay Vora

Trial Role: Chief Investigator

Signature:

A handwritten signature in dark ink, appearing to read 'Ajay Vora', written over a horizontal line.

Date: 01-FEB-2022

This protocol describes the UKALL 2011 trial and provides information about procedures for patients taking part in the UKALL 2011 trial. The protocol should not be used as a guide for treatment of patients not taking part in the UKALL 2011 trial.

AMENDMENTS

The following amendments and/or administrative changes have been made to this protocol since the implementation of the first approved version (1.0):

Protocol Amendment Number	Date	Type	Summary of amendment
8.0	19 th January 2022	Substantial amendment	Clarification of the follow up period throughout the protocol. • 'Minimum of 5 year follow up period' Update to Trial Coordinator details Update to SAE reporting contact details to include email address Re-inserted Figure 6. This had been incorrectly inserted in version 7.0. No changes made to the content. Spelling and formatting corrected throughout.
7.0	21 st February 2018	Substantial amendment	Protocol amended throughout to remove the Randomised Short dexamethasone (R1) induction treatment phase. Update to trial design section – Clarification of standard induction therapy for registered patients Update to abbreviations list (R2 QuEST/TRECA). Update to patient population section to include a trial recruitment end date. 3.3 - Updated given closure of R1- removal of maximum dose of 3 days pre randomisation treatment. Removal of figure 1. 3.4.2 - Update to Figure 2 & Figure 4- to take into account of R1 closure. 3.7.1 ALL/LBL – Updated guidance on IT MTX administration for prevalence of CNS disease at diagnosis. 7.2 - Updated treatment guidelines, for induction and consolidation as per local practice. Removal of diurnal/dietary restrictions for 6-mercaptopurine and co-trimoxazole 7.2.2.5 - Addition of guidance for administration of IT

			chemotherapy via Ommaya reservoir. 7.3 - Trial treatment IMP/NIMP tables updated given the change in status of Induction drugs, including addition of soluble dexamethasone. Summary of treatment tables for regimen A, B & C updated given closure of R1 and addition of QuEST quality of life questionnaire time points throughout, in line with QoL questionnaire. 7.9.2- Asparaginase and Dexamethasone PK sub studies recruitment closure note added. 8.1 - Clarification of adverse event reporting for registration patients. 8.1.2.3 - Clarification of monitoring pregnancies for potential Serious Adverse Events. Section 9 - Clarification of data collections forms required for registration patients Inclusion of Five year follow up data points for registered patients 12.3 Addition of relapse definition to outcome measures. Appendix 4 - MRD laboratories contact details updated. Appendix 13 - assessments amended to take include QuEST time points. Appendix 15 - Notification of closure of sub study added (Dexamethasone PK). Appendix 16 - Removal of MTX special. Appendix 26 - Disease at diagnosis updated to include LBL patients. Updated throughout appendix. Addition of new sub-study - Appendix 27 - TRECA (TRials Engagement in Children and Adolescents) Study. Spelling and formatting corrected throughout.
6.0	3 rd April 2017	Substantial Amendment*	*Implementation following Urgent Safety Measure Protocol amended throughout to include changes due to closure of randomisation 1 (R1) on 31-Mar-2017 Updated contact details for:

			- Chief Investigator - Vincristine study lead - Trial Coordinator Updates made to trial design section (Registration) 4.2 trial entry and exclusion criteria (4.2.1) updated to include closure of randomisation 1 R1 and details of registration of patients on to UKALL 2011 and their possible future inclusion on to the second randomised part of the trial (R2) - Registered patients should receive the standard induction therapy with dexamethasone 6mg/m²/day orally for 28 days - A minimum data set will be collected for registered patients - Registered patients are eligible to participate in cell banking, cytogenetic , Quality of life study and other biological studies (subject to consent) 6.2 Emergency registrations and randomisations-contact details updated
5.0a	8 th August 2016	Non-substantial amendment	7.6.3 Co-trimoxazole (trimethoprim and sulphamethoxazole) re-added to induction treatment Regimen B (removed in error from version 5.0) 7.12.1 Correction made to pre randomisation steroid exposure stating no more than 3 days (previously 7)- As per approved change in protocol version 5.0 Appendix 22 Correction made to pre randomisation steroid exposure stating no more than 3 days (previously 7)- As per approved change in protocol version 5.0
5.0	6 th June 2016	Substantial amendment	Change of Chief Investigator to Professor Ajay Vora Update of funder's name to Bloodwise Updated contact details for Co-investigators and Trial Statistician Change to exclusion criteria: • patients with ALL who due to clinical urgency have received glucocorticoid (dexamethasone or prednisolone) for no more than 3 days. (reduced from 7 days) • patients with NHL or lymphomatous presentation of T-ALL who due to concerns over respiratory compromise or thoracic outlet obstruction have received emergency cytoreduction with glucocorticoid (dexamethasone or prednisolone) for no more than 3 days and/or up to 300mg/m² cyclophosphamide in the

			previous 3 days (reduced from 7 days). Patients diagnosed with mature B-ALL (<u>Burkitt-like, C-MYC re-arranged regardless of morphology or phenotype</u>). Patients with this disease should be treated <u>as mature-B ALL</u> on a suitable protocol for this condition. Registration and subsequent R2 entry option for patients who: • meet all eligibility criteria for R1 except for prior therapy • meet all the eligibility criteria for R1 but who decline entry in to R1 - Registered patients should receive the standard induction therapy with dexamethasone 6mg/m²/day orally for 28 days - A minimum data set will be collected for registered patients - Registered patients are eligible to participate in cell banking, cytogenetic and other biological studies (subject to consent) Clarification of the events included in the primary outcome measure. Event free and overall survival, relapse rate, treatment related mortality and morbidity compared with UKALL 2003 added as new Secondary outcome measure Co-trimoxazole, trimethoprim and sulphamethoxazole doses over 1.0m² amended. Section 3.7.5: Induction Failure Guidance updated following the review of UKALL2003 data. Section 5.1- introduction of a Clinical Update Form for MRD Laboratories. Instructions on when to complete and where to send the form. Section 7.1 and 7.2: clarification that country specific guidance may be followed Section 7.3: • all trial drugs for registered patients will be classified as Non-Investigational Medicinal Products (NIMPs) • addition of cytarabine and hydrocortisone as IMPs when used in place of IMP intrathecal methotrexate Section 7.5.3: patients on Regimen A may receive day 8 intrathecal methotrexate with a day 15 bone marrow (if appropriate) Section 7.5.7: clarification that the folinic acid dose should not be capped Sections 7.5.9, 7.6.9, 7.7.9: clarification that 'no allowances
--	--	--	--

			are made for stopping treatment after the start of interim maintenance' Section 7.12.1: dexamethasone dose modification amended to reflect reduction in number of permissible treatment days prior to randomisation Section 7.13: dose modifications for toxicity updated for high dose methotrexate for protocol M/M-A Section 8.1.2.1 List of Expected SAEs updated to include: • Admissions for fractures • Admissions for post lumbar puncture pain or headache Admission for simple uncomplicated seizure Section 8.1 – AE reporting requirements updated to include registered patients Section 9.1 CRF table removed as data collection is via eRDC Data collection registered patients added Appendix 5: Updated post induction treatment for DS patients. MRD low risk patients should not receive pulses during maintenance. Appendix 6: avoid concomitant use of Nitrous Oxide Appendix 7: addition of further methotrexate guidelines Appendix 12: additional extra blood sample taken at induction on day 9 for Asparaginase substudy. Appendix 13: addition of the Quality of Life Evaluation of Steroids (QuEST) to the Quality of Life study Appendix 14: closure of the MRD flow study. All references about sample collection and timepoints removed. Appendix 15: clarification of time-points and updated telephone numbers within the Dexamethasone Pharmacokinetic Study Appendix 16 IMP supplies updated: a licensed formulation of methotrexate oral suspension is now available Appendix 20: details of genomics sub-study updated - clarification of samples required Appendix 24: updated telephone numbers
--	--	--	--

			Appendix 25: updated flow diagram Appendix 26: addition of an optional neurotoxicity sub-study Spelling errors corrected throughout.
4.0	21 st Dec 2015	Substantial amendment *	*Implementation of the following Urgent Safety Measure • MRD threshold changed for cytogenetic good risk patients • Definition of cytogenetic good and intermediate risk provided for above stratification Screening for ABL class fusion abnormalities in sub-groups of slow responders • Added the following exclusion criteria for R2 based on the above changes: 'B-lineage ALL patients with MRD >1% at day 29 of induction and ABL class fusion abnormalities'
3.0	1 st Oct 2013	Substantial amendment	• Change in trial coordinator • Change from NHL coordinators to LBL coordinators • Amendment to substudy coordinator details • Addition of Vincristine PK Study information • Clarification of the timing of consent • Clarification of recording height at diagnosis • Correction of Appendix references • Addition of prednisolone as an IMP • Correction in length of treatment • Alternative therapy to Allopurinol added • Removal of Thiopurine substudy information • Amendment to Asparaginase substudy sample timings • Clarified Mercaptopurine dosage information • Clarification of BFM consolidation week 6 • Grade 3 and 4 focussed toxicity reporting • Febrile Neutropenia no longer reported as an SAE • Clarification of the term myelosuppression • Addition of events considered to be an expected SAE • Amendment to the DMC meeting schedule • Addition of PTCs and respective MRD labs • Amendment to MRD lab contact details • Amendment to Appendix 12: Asparaginase substudy details • Amendment to Appendix 20: Cytogenetics substudy details • Addition to Appendix 21: TPMT and 6MP details • Addition of Appendix 24: Vincristine substudy details • Amendment to Flow MRD sample distribution • Addition of potential interactions for Methotrexate
2.0	1 st Nov 2012	Substantial amendment	• Change in statistician

			• Change to the exclusion criteria • Change in treatment pathway for lymphoblastic lymphoma (LBL) patients (split into B-cell precursor and T-cell patients) and addition of the rationale for doing this • Addition to reference list (no. 46) • Change in treatment duration for male LBL patients • Clarification added regarding split induction dexamethasone dosing for Down syndrome patients aged 10 years and above • Clarification added about the collection of diagnostic samples from patients with LBL • Addition of information on target dose achievement and alternate day tablet dosing • Amendment to policy on vomited doses • Guidance added on administration of intrathecal methotrexate • Oral mercaptopurine suspension changed from a unlicensed to licensed product • Clarification added regarding continuity of mercaptopurine therapy between treatment phases • Definition of child added as being any patient aged under 18 years • Change in the sample requirements, timings and aims of the dexamethasone sub-study • Clarification on the expected completion times for QoL questionnaires • Cycle duration time added • Bolus does administration of daunorubicin and doxorubicin added • Addition of information on mandatory tests in patients in whom clinical condition precludes bone marrow aspirate • Clarification added regarding events that do not require expedited reporting • Addition of events that do not require reporting on an SAE Form • Clarification of reporting low grade AEs (grade 3 and 4 only to be reported) • Clarification of SAE reporting for patients that are not randomised to R2 • Change in supplier of glucarpidase • Change to the eligibility criteria for completion of Quality of Life (QoL) questionnaires • Change to the timing of QoL questionnaire 5 • Addition of a sampling and analysis centre for the Flow MRD sub-study • Further information added to Appendix 22: T-cell NHL emergencies • Addition of Appendix 23: Diagnostic Flow Cytometry in UKALL 2011
--	--	--	--

TRIAL SYNOPSIS

Title

United Kingdom National Randomised Trial for Children and Young Adults with Acute Lymphoblastic Leukaemia and Lymphoma 2011 (UKALL 2011).

Trial Design

The UKALL 2011 trial is a multicentre, phase III, randomised controlled trial.

On the 31st March 2017 the Data Monitoring Committee (DMC) recommended the closure of randomisation 1 (R1). On analysis of the data available to them on the 15 March 2017, the DMC confirmed there was no clear benefit in administering a short course of dexamethasone in reducing adverse events compared to adverse events experienced on standard dexamethasone. A futility analysis was conducted for the primary endpoint which confirmed the randomisation (R1) to be futile.

As of the 31st March 2017 trial entry in to R1 is no longer permissible. Eligible patients may still be registered in to the trial. Registered patients:

- should receive standard induction therapy
- a minimum data set will be collected for registered patients
- registered patients are eligible to participate in cell banking, cytogenetic, Quality of life study and other biological studies (subject to consent)

Randomisation 2 (R2) will continue as described.

Patients randomised prior to the 31st March 2017 will continue to receive their allocated treatment as defined in the protocol.

Aims

To define whether further refinement of minimal residual disease (MRD) based risk stratification and treatment regimen improves survival whilst reducing overall burden of therapy in children and young adults suffering from acute lymphoblastic leukaemia or lymphoblastic lymphoma (T-cell non-Hodgkin's Lymphoma (NHL) or SmIg-ve precursor B-NHL).

Objectives

Randomised

1. To reduce toxicity through introduction of a short 14-day course of high dose dexamethasone in lieu of the conventional lower dose given for 28 days in induction. (Please refer to trial design section above).
2. To provide more effective Central Nervous System (CNS) prophylaxis and reduce burden of therapy through introduction of high dose methotrexate, and by omission of vincristine and dexamethasone pulses and continuing intrathecal therapy in maintenance.

Non – randomised

3. To decrease toxicity and reduce burden of therapy by administering a single delayed intensification to all patients and limiting augmented therapy to those who are not MRD Low Risk. (*Data collected for all patients up until the closure of R1 31-Mar-2017*).

Outcome Measures

Primary outcome measures

1. Dexamethasone Randomisation (1st Randomisation, R1) .(Please refer to Trial Design section above)

Induction steroid-induced morbidity and mortality defined as all serious, grade 3 or 4 adverse events related to induction and categorised as steroid related or steroid contributory and all induction treatment related deaths during induction.

2. Methotrexate Randomisation (2nd Randomisation, R2)

Central nervous system (CNS) relapse, defined as any relapse with CNS involvement, including combined.

3. Pulses Randomisation (2nd Randomisation, R2)

Bone marrow relapse, defined as any relapse with bone marrow involvement, including combined, Quality of Life measured by PedsQL.

Any event defined as relapse, secondary tumour or death from any cause is also a primary outcome measure for each randomised comparison and the trial overall.

Secondary outcome measures

1. Dexamethasone Randomisation (R1)

Rate of remission, event free & overall survival.

2. Methotrexate Randomisation (R2)

Event free and overall survival, Quality of Life measured by PedsQL, treatment related mortality and morbidity.

3. Pulses Randomisation (R2)

Event free and overall survival, treatment related mortality and morbidity, local relapse (LBL). R2 is a factorial randomisation.

4. Overall trial population

Event free and overall survival, relapse rate, treatment related mortality and morbidity compared to the results from the UKALL 2003 trial.

Patient Population

UKALL 2011 is open to all patients from age 1 (first birthday) to 24 years 364 days (at time of diagnosis) with a first diagnosis of acute lymphoblastic leukaemia or lymphoblastic lymphoma (T-NHL or Smlg negative precursor B-NHL) diagnosed using standard criteria.

Sample Size

Recruitment will continue up until 31-Dec-2018, a minimum of 2640 patients will be recruited to participate in the trial

Main Inclusion and Exclusion Criteria

Inclusion Criteria

UKALL 2011 is open to all patients from age 1 (first birthday) to 24 years 364 days (at time of diagnosis) with a first diagnosis of acute lymphoblastic leukaemia or lymphoblastic lymphoma (T-NHL or Smlg negative precursor B-NHL) diagnosed using standard criteria. Written informed consent is required for all patients and a negative pregnancy test within 2 weeks prior to starting treatment for female patients of childbearing potential.

Exclusion Criteria

The following patients are excluded from entering the trial

1. Infants less than a year old **at diagnosis**. It is recommended that these patients be entered onto the relevant Interfant ALL study.
2. Patients diagnosed with mature B-ALL (Burkitt-like, t(8;14), or C-MYC re-arranged regardless of morphology or phenotype). Patients with this disease should be treated as mature-B-ALL on a suitable protocol for this condition.
3. Patients diagnosed with Philadelphia-positive ALL (t(9;22) or BCR/ABL positive). If randomised patients are subsequently found to have Philadelphia-positive ALL they will be withdrawn from the UKALL 2011 protocol treatment and transferred to a suitable alternative protocol for further therapy.
4. Patients in whom written informed consent has not been obtained from parents and/or patients prior to trial entry.
5. Patients who are sexually active and are unwilling to use adequate contraception during therapy and for one month after last trial treatment.

All patients must be registered within 7 days of starting induction treatment.

Some patients are not eligible for the R2 randomisation. See section 4.1.2 for additional exclusion criteria for R2.

Trial Duration

The recruitment period is expected to last approximately six years with a minimum of five years follow-up following completion of maintenance treatment.

Treatment Summary

Please also see Trial Schema on page 28 for a diagrammatic summary of treatment arms.

INDUCTION THERAPY FOR REGISTERED PATIENTS

Patients with B cell precursor ALL (BCP ALL) should receive standard NCI risk assessed induction in line with national practice

NCI Standard Risk: Patients aged ≥ 1 year and < 10 years old at diagnosis and with a highest white cell count (WCC) before starting treatment of $< 50 \times 10^9/L$.
Patients in this group receive a 3-drug (dexamethasone, vincristine and asparaginase) induction (**Regimen A Induction**).

NCI High Risk: Patients aged ≥ 10 years old at diagnosis, and/or with a diagnostic WCC of $\geq 50 \times 10^9/L$.
Patients in this group receive a 4-drug (dexamethasone, vincristine, asparaginase and daunorubicin) induction. (**Regimen B Induction**).

All Patients with T cell ALL (independent of NCI risk) or Lymphoblastic Lymphoma (LBL) receive induction treatment as per the NCI High Risk induction regimen (Regimen B Induction**).**

All patients with Down syndrome (independent of NCI risk) receive Regimen A induction.

Patients known to have high risk cytogenetics at the start of treatment receive Regimen B induction.

A small number of NCI Standard Risk BCP ALL patients initially treated according to Regimen A induction will subsequently be found to have high risk cytogenetics (MLL (KMT2A) rearrangement, near haploidy, low hypodiploidy, iAMP21, and t(17;19)) and should transfer to Regimen C induction on day 15 of induction. See section 3.3.1 for further details.

NCI High Risk BCP ALL Down syndrome patients and all T-ALL Down syndrome patients with a slow early response as assessed by morphology should also transfer to Regimen C induction on day 15. See section 3.3.2 for further details.

POST-INDUCTION THERAPY (all patients)

Patients with Acute Lymphoblastic Leukaemia (ALL)

All patients with ALL are tested for Minimal Residual Disease (MRD) at day 29 of induction. Post-induction treatment is then stratified on the result of this test as follows:

MRD Low Risk: **MRD <0.005% (cytogenetic intermediate risk); or MRD <0.1% (cytogenetic good risk) at day 29 of induction.** See section 1.2.8 for definition of cytogenetic risk groups.

Patients receive standard NCI directed consolidation therapy – **Regimen A or Regimen B** as per the treatment arm previously assigned for induction.

MRD Risk: **MRD ≥0.005% (cytogenetic intermediate risk); or MRD ≥0.1% (cytogenetic good risk) at day 29 of induction.** See section 1.2.8 for definition of cytogenetic risk groups.

Patients receive augmented Berlin-Frankfurt-Munich (BFM) consolidation (**Regimen C**) and MRD is reassessed upon count recovery from consolidation.

- Patients with post-consolidation MRD ≥0.5% are deemed **MRD High Risk** and are taken off UKALL 2011 protocol treatment.
- Patients with post-consolidation MRD <0.5% are deemed **MRD Intermediate Risk** and continue **Regimen C**.

MRD No Result: **Patients with no MRD result due to inadequate samples or no MRD marker**

Post-induction consolidation therapy in these patients is determined by early response as assessed by morphology:

- Patients with a **slow early response (SER)**, defined as ≥ 25% blasts at day 8 of induction as per Regimen B or day 15 of induction of Regimen A receive augmented BFM consolidation (**Regimen C**). These patients then continue therapy as per Regimen C. No further MRD assessment is required in these patients.
- Patients with a **rapid early response (RER)**, defined as <25% blasts at day 8 of induction as per Regimen B or day 15 of induction of Regimen A continue therapy as per **Regimen A or Regimen B** as per the treatment arm previously assigned for induction. No further MRD assessment is required in these patients.

MRD Low Risk, MRD Intermediate Risk & MRD No Result patients are eligible for the factorial methotrexate and pulses randomisation (R2). MRD Low Risk and MRD No Result RER patients are randomised to receive either standard interim maintenance or high dose methotrexate (Protocol M) followed by a single delayed intensification and either maintenance with pulses or maintenance without pulses (**Regimen A or Regimen B** as per the treatment arm previously assigned for induction).

MRD Intermediate Risk and MRD No Result slow early responder patients will be randomised to receive either Capizzi interim maintenance or high dose methotrexate with asparaginase (Protocol M-A) followed by a single delayed intensification and either maintenance with pulses or maintenance without pulses (**Regimen C**).

Any patient (ALL or LBL) initially treated on Regimen B and found to have high risk cytogenetics should transfer to Regimen C at the beginning of consolidation.

Patients who transferred to Regimen C due to high risk cytogenetics receive Regimen C consolidation and further therapy will be guided by MRD upon recovery from Regimen C consolidation.

Down syndrome patients are not eligible for R2. Post-induction treatment guidelines for these patients can be found in Appendix 5.

Patients with Lymphoblastic Lymphoma (LBL)

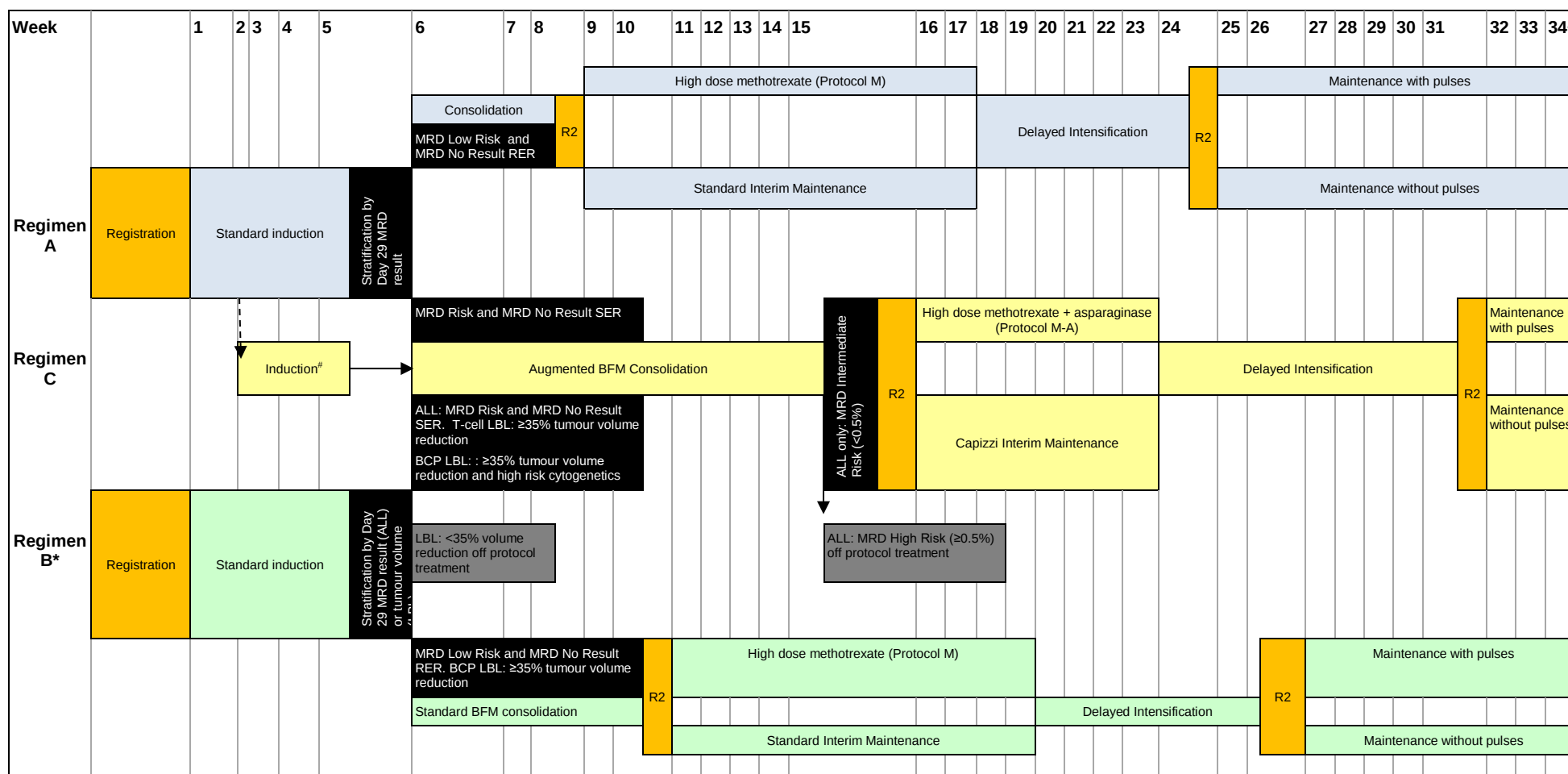
All patients with LBL receive a 4 drug induction (NCI High Risk, Regimen B Induction).

All patients with LBL have assessment of the primary tumour mass as soon as possible after diagnosis and again at the end of induction. Reduction in tumour volume is calculated at the end of induction and treatment then stratified as follows:

Patients with <35% volume reduction after induction therapy are considered poor responders and are taken off protocol treatment. Their care should be discussed with either the Chief Investigator or the LBL coordinators

Patients with ≥35% volume reduction are deemed to be in remission. Patients with T-cell immunophenotype receive augmented BFM consolidation (**Regimen C**), and continue Regimen C thereafter. They are eligible for the factorial methotrexate and pulses randomisation (R2) and are randomised to receive either Capizzi interim maintenance or high dose methotrexate with asparaginase (Protocol M-A) followed by a single delayed intensification and either maintenance with pulses or maintenance without pulses (**Regimen C**). Patients of precursor B-cell immunophenotype receive Regimen B post-induction therapy. These patients are eligible for the factorial methotrexate and pulses randomisation (R2) within Regimen B.

Trial Schema



Induction for BCP ALL patients is directed by NCI risk.

Regimen A induction (NCI Standard Risk) is a 3 drug induction for BCP ALL patients ≥1 year and <10 years and WCC <50x10⁹/L. Down Syndrome patients also receive Regimen A induction treatment.

Regimen B induction (NCI High Risk) is a 4 drug induction for BCP ALL patients ≥ 10 years and/or WCC ≥ 50x10⁹/L and all T-cell ALL and LBL patients.

Post-induction treatment is determined by MRD (ALL patients) or tumour volume assessment (LBL patients). Patients with no MRD result due to an inadequate sample or no MRD marker are assessed by morphology (see section 3.4.2 for details). Patients with slow early response (SER) receive Regimen C treatment following induction therapy. Patients with a rapid early response (RER) receive Regimen A or Regimen B as per the treatment arm assigned for induction.

* All LBL patients and T-cell ALL patients are allocated induction treatment in Regimen B.

[#] Regimen A patients found to have high risk cytogenetics and Down syndrome patients with a slow early response should transfer to Regimen C induction on day 15.

R2 methotrexate and pulses randomisation.

Schedule of Events for ALL patients

Registration																Randomisation 2 (R2)					
	At diagnosis	Day 1 Induction	Day 2 Induction	Day 8 Induction	Day 9 Induction	Day 14 induction	Day 15 Induction	Day 16 Induction	Day 21 induction	Day 28 Induction	Day 29 Induction	Day 30 Induction	End of Induction	Consolidation	End of Consolidation	During Interim Maintenance	End of Interim maintenance	Day 16 DI	During maintenance	18 months	End of treatment
Informed Consent	X												X R2 (MRD Low Risk only)		X R2 (Regimen C only)						
Bone marrow (morphology)	X			X			X (Regimen A only*)														
Bone marrow (MRD)	X										X			X (Regimen C only)	X (Regimen C only)						
Cytogenetics (see 5.1)	X																				
Lumbar puncture	X																				
Asparaginase study samples (see Appendix 12) <u>Closed to recruitment 25-Jul-2017</u>	X				X			X				X		X (Regimen C only)	X (Regimen C only)	X (Regimen C only)		X	X		
Quality of Life Questionnaire & QuEST (see Appendix 13)	X												X				X			X	X
Dexamethasone study samples (SEE APPENDIX 15 Appendix 15 – Dexamethasone pharmacokinetic study - CLOSED) <u>Closed to recruitment 11-Jan-2018</u>		X				X [#]			X ^{**}	X ^{###}											
Vincristine study samples (see Appendix 24) <u>Closed to recruitment 31-Mar-2018</u>			X																		
Chemotherapy associated	X*													X*			X*		X*		X*

Neurotoxicity Sub-study (see Appendix 26)																				
---	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--

The above table provides a summary of sample timings, however the relevant appendices/sections specified should be referred to for full details of samples required/transport.

* Applicable to Neurotoxicity Sub-study group 1 patients only- Group 2 patients study time point refer to appendix 26

Schedule of Events for LBL patients

<u>Registration</u>									<u>Randomisation 2 (R2)</u>					
	At diagnosis	Day 9 Induction	Day 16 Induction	Day 29 Induction	Day 30 Induction	End of Induction	Consolidation	End of Consolidation	During Interim Maintenance	End of interim maintenance	Day 16 DI	During maintenance	18 months	End of treatment
Informed Consent	X					X R2								
Bone marrow assessment	X													
Tumour assessment	X			X										
Cytogenetics	X													
Lumbar puncture	X													
Asparaginase study samples (see Appendix 12) <u>Closed to recruitment 25-Jul-2017</u>	X	X	X		X		X	X	X		X	X		
Quality of Life Questionnaire & QuEST (see Appendix 13)	X					X				X			X	X
Chemotherapy associated Neurotoxicity Sub-study (see Appendix 26)	X*						X*			X*		X*		X**

The above table provides a summary of sample timings; however the relevant appendices/sections specified should be referred to for full details of samples required/transport.

* Applicable to Neurotoxicity sub-study group 1 patients only- Group 2 patients study time point refer to appendix 26

ABBREVIATIONS

AIEOP	Associazione Italiana Ematologia Oncologia Pediatrica (Italian Research Group)
ALL	Acute Lymphoblastic Leukaemia
ANC	Absolute Neutrophil Count
BCP ALL	B-cell Precursor Acute Lymphoblastic Leukaemia
BFM	Berlin-Frankfurt-Munich
BMT	Bone Marrow Transplant
CCG	Children's Cancer Group
CCLG	Children's Cancer and Leukaemia Group
CNS	Central Nervous System
COG	Children's Oncology Group
CR	Complete Remission
CRCTU	Cancer Research UK Clinical Trials Unit
CRF	Case Report Form
CSF	Cerebrospinal Fluid
DI	Delayed Intensification
DIC	Disseminated Intravascular Coagulation
DFCI	Dana Farber Cancer Institute
DMC	Data Monitoring Committee
DMSC	Data Monitoring Safety Committee
DS	Down Syndrome
EDC	Electronic Data Capture
EFS	Event Free Survival
FISH	Fluorescent In-situ Hybridisation
GCP	Good Clinical Practice
GP	General Practitioner
HDMTX	High Dose Methotrexate
HR	High-Risk
HRQoL	Health-Related Quality of Life
ISF	Investigator Site File
LBL	Lymphoblastic Lymphoma
LLR	Leukaemia and Lymphoma Research
LP	Lumbar Puncture
LRCG	Leukaemia Research Cytogenetics Group
MHRA	Medicines for Healthcare products Regulatory Agency
MLL	Mixed Lineage Leukaemia (gene translocation)
6-MP	Mercaptopurine
MRC	Medical Research Council
MRD	Minimal Residual Disease
MTX	Methotrexate
NCI	National Cancer Institute
NHL	Non-Hodgkins Lymphoma
ON	Osteonecrosis
OS	Overall Survival
PCP	Pneumocystis Carinii Pneumonitis

Ph	Philadelphia chromosome
PIS	Patient Information Sheet
QoL	Quality of Life
QuEST	Quality of Life Evaluation on Steroids
R2	Randomisation 2
REC	Research Ethics Committee
RER	Rapid Early Response
RCT	Randomised Clinical Trial
SAE	Serious Adverse Event
SCT	Stem Cell Transplant
SER	Slow Early Response
Smlg	Surface Membrane Immunoglobulin
TMG	Trial Management Group
TRECA	TRials Engagement in Children and Adolescents
TRM	Treatment Related Mortality
TSC	Trial Steering Committee
ULN	Upper Limit of Normal
WCC	White Cell Count

1. BACKGROUND AND RATIONALE

1.1 Background

1.1.1 Role of clinical trials in the treatment of childhood ALL

ALL is the commonest cancer of childhood and affects 420 patients aged 24 years or younger per annum in the UK. Following the first descriptions of curative therapy by the St Jude group in the late 1960s a network of UK clinicians supported by the Medical Research Council (MRC) investigated the benefits of various modifications of treatment through national prospective randomised controlled trials. Accrual to these “UKALL” trials has traditionally been excellent as exemplified by consideration of UKALL 2003 in which more than 97% of eligible patients were recruited over the 7 years of the trial. Consequently, the network has been primarily responsible for standardisation of therapy and uniformity of outcomes across the UK. The trial structure has also facilitated development of the Leukaemia and Lymphoma Research (LLR) funded UK Cytogenetics Database and the national Leukaemia Cell Bank. The UKALL trials have realised a stepwise improvement in prognosis from a 5 year Event Free Survival (EFS) of 35% in 1972 to 87% in 2010. Aspects of the design, approach to risk stratification and outcome of the UKALL trials relevant to UKALL 2011 are now discussed.

1.1.2 UKALL I –XI (1970 – 1997), the age of intensification for all

The UKALL II to VII trials which ran between 1972 and 1979 (n= 1470) failed to demonstrate a significant improvement in the 35% survival seen after multi-agent induction, cranial irradiation and 2 years of continuing therapy in UKALL I. By contrast, UKALL VIII, which recruited during 1980 to 1984, led to a 20% increase in EFS (n=825, 5 year EFS of 52%). This marked improvement was likely due to the policy of continuing therapy without interruption, protracted asparagine depletion and sustained exposure to maximum tolerated doses of continuing therapy. In randomised comparisons UKALL VIII failed to demonstrate a survival advantage for daunorubicin in induction or the use of 3 rather than 2 years of continuation therapy.

The successor trials UKALL X (1985 – 1990, none vs. 1 vs. 2 delayed intensification (DI) courses) tested the benefit of two short 5-day post-remission DI pulsed blocks at week 5 and 20 after induction containing daunorubicin. As the results of UKALL X showed that double DI therapy improved EFS for all patients (EFS 52% (none) v 63% (one) v 72% (two)) this design was taken forward to UKALLXI (1990-97) which tested the value of inclusion of a third seven week BFM style block at week 35. Daunorubicin was omitted from induction due to concerns over cardiotoxicity and the apparent lack of benefit in UKALL VIII. Although UKALL XI did reveal a benefit for the third DI block (69% v 60% 5 year EFS N = 1470) the results in the three DI arms were no better than that seen in the best arm of UKALL X, suggesting that the third DI simply compensated for the omission of induction anthracycline. UKALL XI also tested and found that continuing intrathecal therapy could replace cranial irradiation or high dose methotrexate as CNS directed treatment. All patients received continuing intrathecal therapy as CNS directed therapy and this approach has been adopted in all subsequent UK protocols.

1.1.3 ALL 97 and 97/99 (1997-2002), Stratification by NCI risk and early morphology

ALL 97 (1997-1999) carried forward the three - DI arm of UKALL XI but was modified after it became apparent that the outcome of UKALL X and XI was inferior to that seen in large studies in the US (CCG) and Europe (BFM). Consequently, ALL 97 was modified to a US CCG style regimen. This approach known as ALL 97/99, included changes in the frequency and dosing of asparaginase and allocation of all patients to two CCG modified BFM DI blocks which are less myelosuppressive but provide more prolonged exposure to intensive therapy than the UK type blocks used in UKALL X, XI and 97. For the first time in the UK intensity of therapy was stratified by NCI risk. NCI low risk children (age <10 and WCC <50x10⁹/L received a three drug induction without anthracycline and no systemic consolidation (known as Arm A) whereas high-risk patients received daunorubicin in a four drug induction and a standard BFM consolidation protocol (known as Arm B). Post-induction therapy was also stratified according to early response as defined by morphology early in induction. Slow early responders were allocated intensified therapy (Regimen C) with the so called “augmented BFM protocol (based on increased exposure to vincristine, asparaginase and methotrexate), which had

been shown to confer a 15% improvement in EFS for slow early responders when compared to standard therapy in the US CCG 1882 study.

Both ALL 97 and 97/99 involved a randomised test of the efficacy of dexamethasone 6.5mg/m² x 28 days and prednisolone 40mg/m² for 28 days in induction and for 5 days in monthly pulses during maintenance. A total of 1603 children were randomised and a one third reduction in systemic and CNS relapse was seen with dexamethasone in both phases of the trial. With a 5 year EFS of 80%, ALL 97/99 provided a 12% improvement on that seen in the three DI arms of UKALL XI and a 6% improvement on ALL 97. The main benefit of ALL 97/99 over ALL 97 was a reduction in the rate of relapse involving the CNS, which was seen in both NCI standard and high-risk disease.

1.1.4 UKALL 2003:- Stratification by NCI risk, early morphology and MRD

Although the outcome of the dexamethasone arm of UKALL 97/99 was among the best reported internationally, risk stratification was relatively non-specific. Only 30% of those destined to relapse received the most intensive therapy and consideration of earlier less intensive protocols reveals that many of those cured were exposed to avoidable toxicity. By 2002 several large prospective studies had demonstrated that analysis of minimal residual disease (MRD) was the strongest predictor of outcome in children undergoing identical therapy. Therefore, after successful pilot studies, the first MRD stratified protocol in the UK, UKALL 2003 opened in October 2003. The trial retained the ALL 99 backbone and stratification according to NCI risk and morphologic assessment. All patients received dexamethasone and pegylated *E.Coli* asparaginase (pegaspargase (Oncaspar)) throughout. Rapid early responders (RER) were eligible for a randomised treatment change based on MRD at day 29. Patients with MRD at a level of 0.01% or greater at day 29 received either standard NCI risk-directed therapy (Arm A or B) or augmented BFM (Arm C). Patients with MRD <0.01% at day 29 and a negative MRD result at week 11 (at least one locus quantitative range 10⁻⁴) were randomly allocated a single DI block. All others including children with no MRD result (no sample or no sensitive marker) and those with persistent low-level (positive outside quantitative range of 10⁻⁴, termed, POQR) MRD at week 11 were deemed MRD indeterminate and received standard NCI risk directed therapy with two DI. Interim analysis of UKALL 2003, with follow up to 31st October 2010, reveals a 5 year EFS of 87.7%, and OS of 91.3%. The 5 year EFS for T-ALL at 84% and NCI high risk disease at 83% are particularly encouraging. Importantly, UKALL 2003 has confirmed the predictive value of MRD. A three-fold increase in relapse risk is seen in the MRD high risk group (N=940, 5 year EFS 80.7%) by comparison with children at low MRD risk (N=980, 5 year EFS 95.6%). An intermediate outcome is seen in the MRD indeterminate group (N=937 5 year EFS, 86.7%).

1.1.5 Results of recent clinical trials of treatment of Lymphoblastic Lymphoma

Lymphoblastic lymphoma accounts for 20-25% of all NHL seen in childhood and adolescence. Most are T-NHL and a smaller number represent precursor B (SmIg -ve) – NHL. With modern therapy, survivals of over 70-80% can be achieved but the prognosis of patients who relapse is dismal. The relatively low incidence of LBL in children has largely precluded the commissioning of national disease specific clinical trials and in the UK, as elsewhere; these patients have traditionally been treated with lymphoblastic leukaemia-like protocols. The MRC NHL 9503 protocol recruited 97 patients with lymphoblastic lymphoma between May 1995 and February 2000. Between 1995 and 98 therapy was identical to UKALLXI/ALL 97 with all patients receiving high dose methotrexate (HDMTX) and all eligible for the third intensification block randomisation. Three year survival was 69.4% and OS 79.8%. No benefit was gained from the third intensification block. This outcome was significantly inferior to that seen after treatment according to the NHL-BFM 90 protocol (N=137, 106 T-NHL, 24 pre-B NHL) which had a 5 year EFS of 82%. Therefore, in an attempt to derive more specific information about the optimal therapy of LBL many major European treatment consortia, including the UK MRC adopted the BFM based Euro LB02 regimen. Key elements of this regimen include an anthracycline containing induction, standard BFM consolidation and a single BFM type delayed intensification. In addition all patients received protocol M (high dose methotrexate) as CNS prophylaxis. Pulses of vincristine and steroid were not given in maintenance. Euro LB02 was also designed to examine, by factorial randomisation, the impact of substitution of dexamethasone (10mg/m² for 21 days) for prednisolone (60mg/m² for 21 days) in induction and shortening of maintenance to 18 months.

1.2 Trial Rationale

The results of UKALL 2003 discussed in 1.1.4 represent an improvement in EFS of 6% over ALL 97/99 and are among the best reported to date internationally. UKALL 2011 aims to further improve survival and quality of survival by addressing the four issues apparent in UKALL 2003:

1) Treatment-related mortality and morbidity

The toxicity of UKALL 2003 is unacceptably high in the context of such a high disease free survival. As of October 2010, 100 non relapse deaths were reported in patients enrolled in UKALL 2003; the actuarial risk of treatment related mortality (TRM) in remission is 3.2%. In addition 25% of patients suffered at least one non-haematological SAE. Treatment as per UKALL 2003 also carries a marked reduction in Health Related Quality of Life (QoL). In order to further optimise therapy, UKALL 2011 will examine the impact on relapse and burden of therapy of modifications of the scheduling of dexamethasone in induction, the duration of DI and approach to CNS directed and continuing therapy. Similar concerns over toxicity were apparent in international patients receiving Euro LB 2002 where excess treatment related death in the dexamethasone arm led to early closure of the trial in 2009.

2) Poor prognosis of CNS relapse

In line with international experience UKALL 97/99 and 2003 have shown that increased intensity of systemic chemotherapy combined with protracted intrathecal chemotherapy obviates the need for prophylactic cranial irradiation in nearly all patients. Indeed in UKALL 2003, in which cranial radiation is restricted to the 2% of patients with overt CNS disease (CNS3) at diagnosis, the incidence of any relapse involving the CNS is 4% and that of isolated relapse 2.3%. However, the outlook for children who suffer relapse involving the CNS on UKALL 2003 is likely to be poor with a 5 year EFS of only 30% for these patients relapsing from the similar ALL 97/99 protocol. Moreover it appears that patients suffering an early isolated CNS relapse will only be cured by allogeneic stem cell transplant (SCT). This finding, combined with the burden of therapy of the current approach to CNS prophylaxis justifies examination of an alternative approach in UKALL 2011.

3) Poor prognosis of very early marrow relapse

Although uncommon (2.7% of all patients), very early marrow relapse during therapy (within 18 months of diagnosis) as per UKALL 2003 carries a dismal prognosis with less than 20% survival, even after SCT in second remission. Similar experience is reported by COG and BFM. Improving the prognosis of this group is now key to improving overall survival in ALL. The BFM 2000 trial demonstrated that a proportion of these very high risk children can be identified in first remission by detection of persistently high-level MRD ($>0.5\%$) after 11 weeks of therapy, as per BFM 2000. In UKALL 2011, patients with persistent high level MRD will come off UKALL 2011 protocol treatment. Entry into the primary refractory arm of the national relapse protocol, UKALLR3 which offers novel re-induction and standardised SCT regimen will be considered in such cases.

4) Superior outcome for young adults treated on paediatric protocols.

There is now substantial evidence that young adults with Philadelphia (Ph) negative ALL can achieve greater than 65% EFS without BMT in the majority of patients when treated with a paediatric protocol. In recognition of these data, the upper age limit of UKALL 2003 was increased from 18 to 20 years in 2006 and to 24 years in July 2008. To date 120 patients in the 16 – 24 year group have been enrolled to UKALL 2003. The EFS of 83.8% achieved at 3 years for this group is similar to that for the 10 – 15 year age group. Consequently the upper age limit of UKALL 2011 will be 24 years 364 days and the lower age limit of the new adult ALL trial, UKALL 14, will be 25 years.

UKALL 2003 closed in June 2011. Refinements of molecular MRD technology during UKALL 2003 were such that MRD-stratified therapy based on at least one marker with a quantitative range of 10^{-4} is now applicable to over 93% of patients. In August 2009 the LTSC agreed with the UKALL 2003 coordinators recommendation that the MRD low-risk randomisation should close as recruitment had reached target and there was no excess of relapse in patients randomised to treatment reduction. The high risk randomisation remained open until the closure of the trial. Thus, as with trials conducted in the US and Europe, UKALL 2003 provided proof of principle of the feasibility, wide applicability and

clinical value of MRD-based stratification of therapy in multi-centre randomised controlled trials. Importantly UKALL 2003 has also provided sequential data about health-related quality of life that can now be employed to compare the relative impact of treatment elements in future trials. These data as well as findings of recent international studies, and the ongoing concerns over treatment-related toxicity, CNS and early marrow relapse, outlined above mean that it is an opportune time to open a successor study with the aim of further refining MRD stratified therapy. At present no successor trial to Euro LB 02 is open for the treatment of LBL in the UK and thus there is no mechanism to further refine therapy and monitor toxicity of treatment. Given that UKALL 2011 contains each of the main elements thought to be central to successful therapy of LBL then as in the past, inclusion of lymphoblastic lymphoma patients in the national ALL protocol UKALL 2011 is appropriate.

1.2.1 Treatment stratification

Stratification of Induction according to NCI risk in BCP ALL

Several large trials (ALL 97, CCG 1922 and BFM 2000) have reported a decreased risk of relapse associated with the use of dexamethasone rather than prednisolone during induction. However there are concerns that this benefit may be offset by excess toxicity when anthracycline is used in a dexamethasone based induction. The simple CCG/UK strategy of restricting anthracycline in induction to NCI high-risk cases targets the most toxic induction regimen to those at highest risk of early relapse. Ongoing use of an anthracycline containing four drug induction in NCI high risk BCP ALL can be justified for three reasons. First, in ALL 97/99 and 2003 NCI high risk patients had twice the risk of early relapse compared to those with NCI standard risk disease. Second, in ALL 97 the benefit of dexamethasone over prednisolone was most marked in NCI high-risk patients. Third, results of UKALL 2003 show that almost all NCI high risk patients who are MRD low risk after Arm B induction can be cured with standard therapy (regimen B) and a single DI (N=370 5 year EFS 96.4%). This should be contrasted with the hemi-augmented regimen (regimen C with one DI) universally employed in this group after a prednisolone induction by COG.

Allocation of All Patients with T-NHL and LBL to an anthracycline based induction (Regimen B Induction)

In ALL 99 and 2003 induction was stratified according to NCI risk in all patients regardless of immunophenotype. However, a recent analysis of UKALL 2003 casts doubt on the value of this strategy in T-ALL. Here 41 of 231 RER T-ALL were NCI SR and received no anthracycline in induction and subsequent therapy according to Regimen A. At 80.1% (SE 6.1) and 85.6% (SE 6.2) respectively, the 5 year EFS and OS of this group is inferior to that seen in 190 NCI HR RER patients receiving anthracycline based induction (regimen B) who had a 5 year EFS and OS of 86.7% (SE 2.9) and 91.2% (SE 2.5%) respectively. In LBL the use of anthracycline in induction is likely to have been one of the main reasons for the superior efficacy of BFM 90 over MRC 9503. Consequently in UKALL 2011 all patients with T-NHL and T-LBL will receive anthracycline in induction.

Precursor B immunophenotype LBL is a rare illness but the prognosis is less good than ALL of similar immunophenotype. It is therefore reasonable to treat with a 4 drug induction (Regimen B) in this disorder as in the Euro LB 02 trial.

Use of a Single Delayed Intensification Block in All Patients

Although the benefits of post-induction intensification have been confirmed in an international meta-analysis of 3700 patients in 7 trials, it is important to recognise that this phase of treatment incurs significant side effects and cost. Each of the DI blocks employed in UKALL 2003 carried a TRM of 0.75% and a grade IV toxicity of 8%. The stronger augmented interim maintenance of Arm C of ALL 99/2003 carries a low TRM but a grade IV toxicity of 15%. Frequent hospital visits combined with marked cytopenia and general malaise during DI and Arm C interim maintenance preclude school attendance and disrupt normal family life for up to one year when two DI blocks are given.

Recently published CCG trials have shown no benefit for a second DI block in morphologic RER treated with NCI risk directed regimens identical to Regimen A and B of UKALL 2003. Moreover, the 5-year EFS of 521 MRD low risk cases randomised between single and double DI in UKALL 2003 is 95% and overall survival 98%. EFS is identical in the single and double DI arms. MRD low risk cases have a similar excellent outcome in recent CCG 1991 (EFS 95%, OS 98% N= 340) and BFM 2000 trials (EFS 95%, OS 98% N= 340). Importantly both UKALL 2003 and BFM 2000 showed that the excellent outcome of the MRD low risk group was independent of NCI risk. One potential concern is that follow up of current MRD based trials is relatively short. This is allayed by results of BFM 90 where only one of 55 MRD low risk children has relapsed with a median follow up of 10 years, suggesting the good prognosis of the MRD low risk group will be maintained.

Allocation of MRD Low Risk ALL to Standard Post Induction Therapy and a Single DI (regimen A or B)

In UKALL 2003 MRD low risk was initially defined as no evidence of MRD of 0.01% or higher at day 29, and MRD negative by week 11. This was revised in August 2009 in light of the fact that in patients with MRD <0.01% at day 29 outcome was excellent (95% EFS) regardless of the subsequent week 11 MRD level and that MRD rarely (3 patients in 1454 tested) increased between day 29 and week 11. Thus from that point MRD low risk was defined on the basis of the day 29 result alone. At the same time it became apparent that patients with day 29 MRD level between 0.01 and 0.005% experienced a relative excess of bone marrow relapse by comparison with those with day 29 MRD below that level. Thus in UKALL 2011 the MRD Low Risk threshold was redefined as no evidence of MRD at a level of 0.005% or greater at day 29 using at least one and optimally two markers with quantitative range of 10^{-4} .

Subsequent analysis in November 2015 by cytogenetic sub-group and MRD demonstrated that there was no difference in overall survival or relapse risk for cytogenetic good risk patients with MRD <0.1% who received Regimen C compared with Regimen A or B (Table 1). Regimen C is associated with significant excess morbidity related to asparaginase, intra-venous methotrexate and vincristine intensification. As such, a decision was made to alter the threshold at which patients with good risk cytogenetics are classified as MRD risk to a level of 0.1% or more at day 29 of induction. Patients with good risk cytogenetics and MRD less than 0.1% were therefore treated as MRD low risk from 3 December 2015.

It should be noted that as detailed above the outcome of NCI standard risk T-ALL treated with Regimen A of UKALL 2003 was inferior to that seen in NCI High Risk patients on Regimen B. Thus NCI Low Risk MRD Low Risk T-ALL will receive standard post induction therapy as per regimen B.

Table 1: Relapse rate (RR) at 5 years for UKALL2003 patients with good risk cytogenetics (N=1311*) stratified by MRD level at the end of induction.

	All Cytogenetics Good Risk patients *		MRD positive (>0.01%) patients randomised to stay on Regimen A/B		MRD positive (>0.01%) patients randomised to stay on Regimen C		Cytogenetics Good Risk patients who received regimen A/B**		Cytogenetics Good Risk patients who received regimen C**	
	N (%)	RR % (95% CI)	N (%)	RR % (95% CI)	N (%)	RR % (95% CI)	N (%)	RR % (95% CI)	N (%)	RR % (95% CI)
0%	416 (32%)	1.64 (0.7-3.6)					414	1.65 (0.7-3.7)		
0-0.005%	387 (30%)	3.98 (2.3-6.9)					386	3.99 (2.3-7.0)		
0.005-0.01%	129 (10%)	3.78 (1.4-9.8)					128	3.81 (1.4-9.9)		
0.01-0.1%	251 (19%)	3.91 (2.0-7.4)	79	2.68 (0.7-10.3)	77	2.83 (0.7-10.9)	159	4.72 (2.3-9.7)	92	2.48 (0.6-9.6)
0.1-1.0%	96 (7%)	11.28 (6.0-20.7)	34	17.9 (7.8-38.1)	31	7.1 (1.8-25.4)	61	14.52 (7.1-28.3)	35	6.29 (1.6-23.0)
1-10%	26 (2%)	8.42 (2.2-29.9)	10	11.1 (1.6-56.7)	8	12.5 (1.9-61.3)	15	7.14 (1.0-40.9)	11	9.1 (1.3-49.2)
>10%	6 (<1%)	20.0 (3.1-79.6)	5	25.0 (3.9-87.2)	1	-	5	25.0 (4.0-87.2)	1	-

* Excludes patients who had a slow early response, induction failure or induction death; ** These columns include randomised patients as well. Four patients have been excluded because they were coded as MRD negative, received regimen C but were not slow early responders.

Allocation of MRD Intermediate Risk ALL patients to Augmented Post Induction Therapy and Single DI (Regimen C)

UKALL 2003 also examined the value of augmented BFM therapy (Regimen C of UKALL 2003) in morphologic RER with MRD at a level of 0.01% or greater at day 29. Five hundred and thirty three patients entered into a randomisation comparing standard treatment (Regimens A or B = standard arm, n = 266) with the more intensive Regimen C (intensification arm, n = 267). Of these, 332 (62%) were NCI SR and 201 (38%) NCI HR. With follow-up to October 2011 (median 3 years 11 months), the 5 year EFS is significantly better for patients in the intensification arm (91%) compared with the standard arms (82%) due to a halving of relapse risk (OR 0.57, 95% CI 0.34 – 0.95, 2p = 0.03). The improved EFS translates into a trend for better overall survival (OR 0.57, 95% CI 0.31 – 1.05, p = 0.07). There was no heterogeneity in benefit of Regimen C for sub-groups defined by NCI risk, immunophenotype or cytogenetics. The risk of death in remission was not significantly different in the two arms (Intensification = 7, Standard = 9, OR 0.76, 2p = 0.6) but there was an excess of SAEs related to asparaginase, intravenous methotrexate with Regimen C.

Further support for the benefit of intensification of therapy in MRD risk groups comes from the St Jude Total XV study in which intensification for those with MRD between 0.01 and 0.99% at day 42 improved 5 year EFS from 43% in previous trials to 79% in Total XV. There is also strong evidence from the BFM 2000 study that sequential MRD analysis during the first few months of therapy can improve prognosis for patients at very high risk of relapse. In this study, children with persistent high level MRD at 12 weeks (a group with a 70% risk of early relapse in the BFM 90 study) were allocated high-risk (HR) blocks and SCT. This strategy was highly effective and outcome for the MRD HR group in BFM 2000 was much improved at 63%. MRD analysis during the HR phase showed that continued cyto-reduction was the crucial determinant of outcome. Those with a good response could be cured with continued chemotherapy although there was an advantage for SCT in this group. Of particular note is the correlation between response to HR therapy and week 12 MRD level:- only 35% of patients with MRD >0.5% at week 12 responded, whereas response was seen in 85% with MRD below that level.

The results of UKALL 2003, St Jude Total XV and BFM 2000 as well as concerns over toxicity of augmented BFM with 2 DI are the basis for the change in post induction therapy for patients with MRD $\geq 0.005\%$ at day 29 of UKALL 2011. In UKALL 2011 these cases will form the MRD Risk group and will all receive augmented BFM (regimen C) consolidation. Following the lead of BFM 2000 MRD will be measured again upon recovery from consolidation. Those with MRD $\geq 0.5\%$ (MRD High Risk) upon recovery from augmented consolidation are very unlikely to be cured with ongoing chemotherapy and entry onto the national relapse refractory ALL trial with novel cyto-reduction followed by SCT will be considered. The remaining MRD Intermediate Risk group will receive augmented BFM therapy with a single DI (Regimen C). The value of this regimen to this group is inferred from Total XV and the non-MRD stratified CCG 1961 and 1991 trials. Both CCG 1991 and 1961 showed a reduction in relapse after intensified interim maintenance but no value for a second DI in RER with NCI risk directed regimens identical to Arm A and B of UKALL 2003. The coordinators acknowledge that an augmented BFM protocol (Arm C) with a single DI phase has not been proven in morphologic SER cases. However, results in UKALL 2003 show 34% of morphologic SER had MRD <0.01% at the end of induction. This Slow Early Response (SER) MRD low risk group has 5 year EFS of 94.9% and is a candidate for less aggressive therapy. It is expected that identification of persistent high-level MRD upon recovery from consolidation will highlight the very small sub-group at very high risk of relapse who may benefit from even more intensive therapy.

Allocation of T-LBL to Augmented Post Induction Therapy and a Single DI (Regimen C) and precursor B-LBL to standard post induction therapy and a single DI (Regimen B)

In T-LBL relapse normally occurs early in treatment and is very difficult to salvage. There is evidence of increased risk of relapse in patients who show a poor response to induction as defined by volumetric assessment with CT scan or ultrasound. In Euro LB 02 and the subsequent UK guideline a good response was defined as a 35% reduction in tumour mass. Poor responders as defined in this way were candidates for experimental therapy. This approach to stratification of post induction therapy will continue in UKALL 2011. Poor responders will come off protocol whilst good responders

will continue with Regimen C. The rationale for the use of Regimen C in LBL is based on the fact that although the outcome for this group is excellent, with a 5 year EFS of over 80%. The prognosis for those who subsequently relapse is dismal. As in ALL there is a need to balance toxicity of therapy with risk of relapse. Attempts to reduce relapse through increased use of anthracycline and cyclophosphamide on a BFM backbone were unsuccessful in CCG 5791. Optimally therapy for LBL should now be guided by biology and sensitive assessment of response. Until such technology is available it is reasonable to consider further intensification for all patients with advanced disease through the use of augmented exposure to vincristine and asparaginase in consolidation, interim maintenance and delayed intensification employed successfully in recent UK and US ALL protocols. Thus all patients with T-LBL who are in remission at the end of induction will be allocated therapy on Regimen C.

Precursor B lineage LBL is uncommon and there is little evidence base to guide management. The outcome of these patients is however inferior to leukaemic children of the same immunophenotype with similar NCI criteria. Those patients with precursor B-LBL who achieve an adequate response by imaging will be given standard post induction therapy and a single DI (Regimen B).

UKALL 2011 will investigate through prospective randomisation:

Randomisation 1 (R1) – dexamethasone randomisation

- 1) In induction, the effect on serious treatment-related toxicity of receiving either a dexamethasone schedule of 10mg/m²/day for a total of 14-days, or the current standard UK schedule of 6mg/m²/day for 28 days. **Closed 31st March 2017 (Please refer to Trial Design for update on closure of R1).**

Randomisation 2 (R2) – methotrexate and pulses randomisation

- 2) In interim maintenance therapy, the effect on CNS relapse risk and quality of life of receiving either high dose methotrexate without prolonged intrathecal therapy or the current standard UK CNS-directed ALL therapy with protracted intrathecal therapy.
- 3) In maintenance therapy, the effect in patients on bone marrow relapse risk and quality of life of receiving monthly pulses of vincristine and dexamethasone.

The methotrexate and pulses randomisation is a factorial design with patients being randomised to receive either high dose methotrexate or standard interim maintenance followed by a single delayed intensification and either maintenance with pulses or without pulses of vincristine and dexamethasone.

The randomisations are discussed further below.

1.2.2 Rationale for Use of Dexamethasone

Over the last twenty years more than 10,000 patients have been enrolled in randomised trials designed to assess the relative efficacy and toxicity of dexamethasone and prednisolone in ALL (reviewed in Nachman, BJH 149 638-652). It is now clear that substitution of dexamethasone for prednisolone at a ratio of 1:6 (as in ALL 97/99, CCG 1922 and BFM 2000) reduces the risk of relapse by up to one third. By contrast the use of higher relative doses of prednisolone (ratios of 1:7.5 (Tokyo L95-14) or 1:10 (EORTC 58951) negates the impact of dexamethasone on relapse. As experience with dexamethasone has increased so have concerns that increased efficacy may be offset by high induction mortality, particularly when an anthracycline is used. These are exemplified by the BFM 2000 study which reported that, following a 7 day prednisolone pre phase, the use of dexamethasone (10mg/m² for 21 days, tapered to day 28) during a four drug (including an anthracycline) induction produced a one third reduction in risk of relapse when compared to prednisolone 60mg/m² given for 21 days, tapered to day 28. However the reduction of relapse seen with dexamethasone was offset by

an increase in treatment related mortality, particularly in patients aged over 10 years in whom induction TRM was 4%. Indeed, sub group analysis of BFM 2000 revealed that in BCP ALL there was no gain in overall survival in the dexamethasone arm, whereas there was a clear improvement in outcome in T-ALL. Consequently, BFM have chosen to continue to use prednisolone at 60mg/m² within an anthracycline containing induction in all patients with BCP ALL in their next protocol.

By contrast with the BFM approach, current and future studies conducted by COG and FRALLE are designed to exploit the reduced risk of relapse seen after dexamethasone in induction in BCP ALL, proven in ALL 97/99 and CCG 1922. Both groups continue to employ the simple strategy of restricting the use of anthracycline in a dexamethasone based induction to NCI high-risk BCP cases thereby targeting the most toxic induction regimen to those at highest risk of early relapse. Moreover at 2.4% the TRM of the NCI high risk induction in UKALL 2003 which employs dexamethasone at 6mg/m² is identical to that seen when prednisolone was used at 60mg/m² in BFM 2000 and COG 0232. A further advantage of NCI risk directed induction is highlighted by the recently published CCG 1991 study which shows that NCI standard risk BCP ALL patients receiving 6mg/m² dexamethasone in a non-anthracycline induction do not benefit from a second DI. No such data exist for patients receiving a prednisolone based non anthracycline induction. Lastly, it is also important to note that the excellent outcome after standard NCI directed therapy (Regimen A or B) and a single DI in the MRD low risk group in UKALL 2003 was derived from the use of a dexamethasone based induction.

Euro LB02 was designed to explore the value of dexamethasone over prednisolone in LBL. Patients received an induction identical to BFM 2000 and were randomly allocated 60mg/m² of prednisolone or 10mg/m² of dexamethasone. The protocol was closed in 2009 due to international patients suffering an excess of significant toxicity. Nevertheless the coordinators of UKALL 2011 believe that it is important to exploit the potential value of dexamethasone in all types of lymphoblastic malignancy in children and adolescents. As discussed above the TRM of the anthracycline containing induction in UKALL 2003 which employs dexamethasone at 6mg/m² was identical to that seen in the prednisolone arm of BFM 2000. Moreover the efficacy of prednisolone 60mg/m² vs. dexamethasone at 6mg/m² are equivalent in ALL. Thus it is reasonable to include all patients with LBL in a protocol designed to define the optimal scheduling of dexamethasone in an anthracycline based induction.

1.2.3 Rationale for Dexamethasone Randomisation

For the reasons outlined in section 1.2.2 UKALL 2011 will employ dexamethasone in all phases of therapy. Initially the trial was designed to explore whether the use of a shorter course of higher dose dexamethasone in induction could reduce toxicity whilst maintaining efficacy. Toxicity was assessed as treatment related mortality and adverse events (AEs) via two types of AE steroid related and steroid contributory (see section 2.2 for definitions). As discussed above the three drug non-anthracycline induction regimen (Regimen A) of UKALL 2003 has a lower TRM than the anthracycline containing dexamethasone arm of BFM 2000 in children under 10 years of age (0.7% v 1.4%). Nevertheless, the regimen carries significant morbidity, including a 4.9% combined incidence of steroid related and steroid contributory SAEs. In line with the experience of other groups, the toxicity of the anthracycline containing four drug induction regimen used in NCI high risk children on ALL 97/99 and 2003 is relatively high with a 2.4% induction TRM and an 8.2% incidence of steroid related and steroid contributory toxicity. By contrast with BFM 2000, no excess TRM was seen in NCI high risk patients over 10 years of age on UKALL 2003. Table 2 shows the toxicity of dexamethasone based induction by regimen in UKALL 2003.

Table 2: Toxicity of dexamethasone based induction in UKALL 2003

	AA	BB/BC/AC	Total
Number Treated	1126	1128	2287
Induction TRM	7	27	34
Induction "Steroid Related" SAE	9	26	35
Induction "Steroid Contributory" SAE	44	67	111

Osteonecrosis*	5	50	55
----------------	---	----	----

Table 2 Toxicity of dexamethasone based induction by regimen in UKALL 2003. AA: patients treated on Regimen A throughout induction, BB: patients treated on Regimen B throughout induction, BC/AC: patients initially treated on Regimen A/B induction and transferred to Regimen C induction. NCI Standard risk RER and no Ph POS, iAMP21 or hypodiploidy received no anthracycline in induction (AA). All others received anthracycline four-drug induction (BB/BC/AC). SAEs are sub categorised as steroid related and steroid contributory (see section 2.2 for definitions). *Incidence of osteonecrosis at any time during therapy (85% occur within 2 years of diagnosis of ALL).

The 14-day course of 10mg/m² dexamethasone selected as the experimental induction arm of UKALL 2011 was designed to maximise early cytoreduction whilst avoiding the infective and other toxicities seen with the 5 week exposure to steroids in the control arm. Efficacy and safety of the experimental induction regimen can be inferred from the UKALL R1 protocol where it was effective (95% CR) and relatively safe (TRM = 1.3 %) when used with epirubicin, vincristine and asparaginase. The potential for more rapid cytoreduction to translate to decreased relapse risk was supported by COG and BFM data which show over a 95% EFS in children with no MRD at a level of 0.01% by day 15 of therapy. The efficacy of induction with 10mg/m² dexamethasone for 14 days versus prednisolone 60mg/m² for 28 days was recently compared in the COG 0232 trial for NCI HR patients treated with a four drug induction and a protocol very similar to Regimen C of UKALL 2003. Preliminary results were presented at ASCO in 2011. There was no excess induction TRM in the dexamethasone arm and the anti-leukaemic efficacy of dexamethasone was superior in patients aged 1-9 years and equivalent in those 10 years and over.

An interim review of the data along with a formal futility analysis by the independent data monitoring committee of UKALL 2011 on 30 March 2017 led to them recommending closure of this randomisation as it was unlikely that the short dexamethasone schedule would be associated with less toxicity than the standard schedule. The results of this analysis will be made available once published.

1.2.4 Rationale for use of split dexamethasone in patients aged 10 and over

The design of optimal therapy for cancer requires a careful balance between efficacy and long term toxicity of therapy. Serial clinical trials in ALL have revealed how best to minimise long term toxicity consequent on exposure to radiotherapy, anthracycline, alkylating agents and topoisomerase inhibitors whilst maintaining EFS. The new challenge is to maintain the excellent outcomes for older patients seen with modern “steroid intensive” regimens whilst reducing the incidence of disability due to osteonecrosis of bone. This complication can have a profound negative impact on long term quality of life, in some cases requiring multiple joint replacements and residual disability. Unfortunately as yet there is little consensus about the natural history or optimal approach to diagnosis and management of osteonecrosis. It is however clear that osteonecrosis is more common in older patients (99 of 110 cases of ON in UKALL 2003 were aged over 10 years at diagnosis) and in protocols with high cumulative steroid exposure.

The COG 0232 protocol, which compared prednisolone 60mg/m² for 28 days with 14 days of dexamethasone 10mg/m² regimen in induction for NCI high risk patients, is of particular relevance to the design of UKALL 2011. All patients in this study received a single augmented DI and one augmented interim maintenance (ie Regimen C of UKALL 2011). In 2008 the DMSC recommended closure of the dexamethasone arm of COG 0232 in patients aged over 10 years because of an excess of ON when compared to that seen in patients receiving 60mg/m² of prednisolone in induction. Following modification of the protocol, to mandate the use of prednisolone in induction and discontinuous dexamethasone in DI in all patients, the incidence of symptomatic ON on COG 0232 is now 16% in patients over 10 at diagnosis. This is very similar to the rate of symptomatic ON seen in patients over 10 years treated on UKALL 2003, all of whom received 28 days of 6mg/m² of dexamethasone in induction.

As discussed above experience in COG 0232 suggests it is unlikely that the use of continuous 14 days of 10mg/m² of dexamethasone for patients aged over 10 years will reduce the incidence of symptomatic ON in patients aged over 10 years enrolled on UKALL 2011. However data from recent COG studies suggests that it may be possible to reduce osteonecrosis but maintain efficacy through

introduction of a discontinuous or split dexamethasone regimen in induction. In CCG 1961 patients were randomised to one DI with 21 days of continuous dexamethasone at 10mg/m² or two DIs with discontinuous dexamethasone 10mg/m² (day 1-7, day 15-22). In this trial the rate of symptomatic osteonecrosis was lower (5% v 10%) in those receiving discontinuous dexamethasone suggesting that sustained exposure to high dose dexamethasone may be key to development of osteonecrosis. Based on this and driven by the desire to continue to harness the increased benefits of a dexamethasone based induction set out above, the experimental arm of the dexamethasone randomisation in UKALL 2011 will employ split dexamethasone in patients aged over 10 years at diagnosis. As the use of split dosing in induction is unproven, clear stopping rules based on day 29 MRD risk and relapse will be provided to the trial DMSC.

Most importantly, as well as assessing the incidence of osteonecrosis by induction regimen the design of UKALL 2011 will provide further information as to which post-induction elements influence osteonecrosis. Patients will be non-randomly allocated ongoing asparaginase during interim maintenance on the basis of MRD and randomly allocated dexamethasone pulses during maintenance. This aspect of the design of UKALL 2011 will also allow comparison with the results of COG 0232 and provide opportunity for add on studies designed to clarify the natural history and risk factors for osteonecrosis at the same time as defining optimal approach to diagnosis and management. The management of osteonecrosis is discussed in Appendix 17.

1.2.5 Rationale for high dose methotrexate randomisation

Concerns over toxicity, particularly secondary brain tumours, have led all of the major treatment consortia to limit the use of cranial radiation as CNS directed therapy in ALL. Increased intensity of systemic chemotherapy combined with protracted intrathecal chemotherapy or high dose intravenous methotrexate can prevent CNS relapse in most children. Thus in UKALL 2003, in which cranial irradiation is reserved for those with overt CNS disease at diagnosis, the incidence of any relapse involving the CNS is now 4% and that of isolated relapse 2.3%.

In spite of this improvement the management, prevention and prediction of CNS relapse presents an ongoing challenge. A recent analysis of UKALL trials between 1985 and 2001 reveals that although overall relapse rates have decreased by 49% the proportional incidence and timing of isolated CNS failure is unchanged. However the prognosis of isolated CNS relapse has changed. Whereas early isolated CNS relapse could be salvaged without resort to SCT in over 70% of children relapsing after treatment as per UKALL XI, outcome in 6 children with early isolated CNS relapse in ALL 97/99 and 2003 who did not receive SCT was dismal and even with SCT EFS is 45% . Of equal concern is the fact that the current UK approach to CNS directed therapy carries a significant burden. Patients undergo up to 26 therapeutic lumbar punctures under general anaesthesia. Finally, although the relative risk of CNS failure parallels other high risk features, MRD is a relatively poor predictor of isolated CNS failure; - 15 of 33 relapses in children with MRD less than 0.005% at day 29 of UKALL 2003 involved the CNS in isolation.

UKALL 2011 will investigate whether administration of intravenous high dose methotrexate (HDMTX) is a superior alternative to the current UK approach to CNS prophylaxis. The primary end point will be a reduction in CNS relapse but strong emphasis will also be placed on impact on HRQoL. HDMTX (total dose 20 grams/m² in 8 weeks) has been used successfully by the BFM for 20 years. The regimen, known as Protocol M, has limited toxicity and provides excellent CNS control without the need for ongoing intrathecal chemotherapy and cranial irradiation in most cases. Moreover there is in vitro evidence that HDMTX may be particularly effective in systemic control of high hyperdiploid BCP disease and T-ALL. The efficacy of HDMTX compared with continuing intrathecal therapy has previously been trialled in the UK, where it was shown to reduce CNS relapse risk from 6.6% to 3.3% in children with presenting WCC <50 x10⁹ /L.

Use of Protocol M in MRD Low Risk ALL

Support for the use of intravenous MTX to reduce CNS relapse in low risk patients comes from CCG 1991. In that study escalating Capizzi methotrexate (without folinic acid rescue) and 10 additional

doses of vincristine were used with all patients receiving continued intrathecal therapy and pulses in maintenance. UKALL 2011 will investigate whether the use of Protocol M alone in lieu of standard interim maintenance reduces the relative excess of CNS relapse and burden of therapy in MRD Low Risk patients. Patients with T-ALL and a WCC $>100 \times 10^9/L$ who receive Protocol M will also receive continued intrathecal therapy as practised by AIEOP. No patient in this group will receive cranial radiation.

Use of Protocol M-A in MRD Intermediate Risk ALL

CCG 1961 demonstrated a reduction in marrow relapse after augmented interim maintenance and DI in NCI high risk RER (see 3.4). No reduction in CNS relapse was seen: the relative excess of CNS relapse is recognised as an ongoing issue by COG. It is reasonable to suggest, therefore, that exposure to higher systemic doses of methotrexate may reduce CNS relapse in this group. This was tested in the COG 0232 protocol in which BCP NCI HR patients were randomised to receive either dexamethasone or prednisolone in induction (see section 1.2.1) and then either Capizzi or HD MTX (as Protocol M) as interim maintenance. Preliminary data from this study shows that the Dexamethasone HD MTX combination was the mostly effective in patients aged 1-9 – 5 year EFS 93.5%. Whilst comparison with Protocol M alone as in COG 0232 is a reasonable option, data from DFCI as well as CCG suggest that protracted asparagine depletion is also important in higher risk ALL. Administration of HDMTX with asparaginase was trialled as Protocol M-A in BFM 90. Here patients were randomly allocated to a regime of 25,000 units of erwinase 48 hours after each HDMTX infusion in attempt to maximise intracellular toxicity of HDMTX. Although no excess of toxicity was seen this approach did not lead to increased efficacy when randomly compared to HDMTX alone. An alternative strategy is to attempt to achieve more protracted asparagine depletion during protocol M. Thus in UKALL 2011 protocol M-A will deliver Asparaginase PEG at day 3 and 23 of Protocol M, thereby providing a true comparison with the current standard Capizzi approach. A similar regimen is currently being used by the NOPHO 2008 study with no excess TRM in over 400 patients treated to date. In summary UKALL 2011 will investigate whether the use of HDMTX and asparaginase in lieu of augmented BFM interim maintenance reduces CNS relapse risk and burden of therapy in MRD high risk patients. Patients with T-ALL and a high WCC $>100 \times 10^9/L$ who receive Protocol M or M-A will also receive continued intrathecal therapy in line with the use of extra CNS prophylaxis used in this group by AIEOP/BFM. No patient in this group will receive cranial radiation.

Use of Protocol M-A in Advanced Stage LBL

Until recently, protocols based on the BFM ALL backbone represented the most successful approaches to the treatment of LBL. Data from the NHL BFM 90 and 95 trials reveals that CNS negative LBL with a good response to induction do not suffer an excess of CNS relapse when HDMTX, as protocol M, is substituted for cranial irradiation as CNS prophylaxis. The 5 year EFS of this group in NHL-BFM 95 (n=156) was 82% (Standard Error 3%). The value of Protocol M in LBL has recently been questioned by the results of CCG 5971 in which CNS negative LBL patients received a regimen identical to BFM 95 but protocol M was randomly replaced by oral interim maintenance and continued intrathecal therapy. All patients received vincristine and steroid pulses in maintenance. Preliminary data from this study revealed no difference in EFS in patients receiving oral interim maintenance and continuing intrathecal, who had an 84% EFS at 5 years (n=257). On the basis of the above it is clear that good survivals can be achieved in CNS negative LBL without recourse to cranial irradiation but that, as in ALL, the relative role of intravenous methotrexate or continuing intrathecal therapy as CNS therapy is unknown. It is therefore reasonable to include LBL patients in the Methotrexate randomisation in UKALL 2011. It is also clear that there remains room to further improve survival for this group of patients and thus it would seem appropriate to maximise exposure to asparaginase through either the relatively simple outpatient based Capizzi and continuing intrathecal approach or the in-patient based protocol M-A.

1.2.6 Rationale for Pulses randomisation

Intensification of maintenance through administration of vincristine and steroid “pulses” has been widely used since the 1970s. Historically the CCG trials, which form the basis of current UK regimens, have used pulses and in UKALL 2003 all patients received up to three years of monthly pulses of intravenous vincristine (1.5mg/m², max 2mg) and 5 days of oral dexamethasone (6mg/m²). Whilst specific assessment of the toxicity of pulses was not part of UKALL 2003, it is clear from discussion

with patients, parents and clinical teams that they are a source of significant morbidity and burden of care.

The primary aim of the administration of pulses is a reduction in marrow relapse but there is data from earlier trials, notably CCG 161 that pulses may also reduce the incidence of relapse involving the CNS. In 1996 a meta-analysis which included four randomised trials of pulses with over 1000 entrants showed that pulses conferred a 10% increase in 5 year EFS from 59% to 69%. However, at 69%, the 5 year EFS in the pulses arm of the 1996 meta-analysis is relatively poor by modern standards. Indeed, a recently published update of the 1996 meta-analysis suggests that the gain in terms of relapse risk from pulses is limited to earlier less intensive protocols. UKALL 2011 will therefore examine the role of pulses both in patients receiving continuing intrathecal therapy and those receiving Protocol M.

Omission of pulses in MRD Low Risk

In this factorial design 25% of the MRD low risk group will receive an experimental regimen with neither pulses nor HDMTX. They will receive continuing intrathecal as given in UKALL 2003. Even if this change results in a 50% increase in marrow relapse, this will mean an increase in absolute risk from 1.5% to 2.25%. Moreover it is likely excess relapses will be late and likely salvageable. If this least intensive arm is successful UKALL 2011 will have been able to identify a group of 33% of patients who can be cured with one DI, no pulses and no intravenous MTX. If it is not, the factorial randomisation and quality of life questions will allow us to define the optimal approach in this low risk group.

Omission of pulses in MRD Intermediate Risk

Again, as a consequence of the factorial design 25% of MRD intermediate risk patients will receive an experimental regimen involving a single augmented interim maintenance and a single DI with no pulses. They will receive continuing intrathecal as given in UKALL 2003. Data from the high risk randomisation of UKALL 2003 suggests that NCI SR patients with MRD >0.01% at day 29 treated on Regimen C have a 93.8% (89.7-97.9%) 5 year EFS, whereas NCI HR patients with MRD >0.01% at day 29 have a 84% (75.7% -92.9%) 5 year EFS. Given the additional methotrexate and asparaginase related toxicity of Regimen C documented in UKALL 2003 as well as meta analyses showing that vincristine and dexamethasone pulses do not increase anti-leukaemic efficacy in patients exposed to intensive early therapy it is reasonable to test the balance between anti-leukaemic efficacy and toxicity of pulses (including impact on HRQoL) in the MRD intermediate risk cohort in UKALL 2011. The outcome for this group will be closely monitored by the DMC in order to detect any excess risk of relapse.

Omission of pulses in Advanced Stage LBL

In LBL BFM protocols have not employed pulses whereas all patients received monthly vincristine and prednisolone pulses in COG 5971. UKALL 2011 will attempt to define the role of pulses after intravenous methotrexate based interim maintenance in lymphoblastic leukaemia and by inference in LBL. As a consequence of the factorial design 25% of LBL patients will receive an experimental regimen involving Capizzi MTX and DI with no pulses. They will receive continuing intrathecal as given in UKALL 2003. It is argued that this decision can be justified in that, by comparison with NHL BFM 95 and Euro LB02 these patients will receive increased exposure to vincristine and asparaginase in augmented consolidation and delayed intensification as well as additional vincristine within the Capizzi interim maintenance block.

1.2.7 High Risk Cytogenetics in B-cell precursor ALL (See also appendix 25 for information on testing for ABL-class fusion abnormalities)

Over the last 20 years the Leukaemia Research Cytogenetics Group (LRCG) has identified a number of relatively uncommon cytogenetic abnormalities that have correlated with a poor outcome. Based on data from ALL 99 and 2003 five high risk cytogenetic abnormalities are recognised in UKALL 2011:- iAMP21, t(17;19)(q22;p13), MLL (KMT2A) rearrangement, near haploidy and low hypodiploidy. Although several trials have investigated the value of SCT in 1st complete remission (CR1) for these patients there is no randomised study that proves the superiority of SCT over response directed chemotherapy. Thus, in UKALL 2011 patients with high-risk cytogenetics will receive a four drug induction followed by Regimen C augmented consolidation. Further therapy will then be directed by post-consolidation MRD. This section presents a brief rationale for the recommended approach.

Intrachromosomal amplification of chromosome 21 (iAMP21)

Analysis of ALL97 indicates that iAMP21, seen by cytogenetics as dup (21q), with amplification of the RUNX1 (AML1) gene detected by FISH was associated with a poor prognosis. Outcome for this group was much improved in UKALL 2003 after adopting a policy in which these patients were allocated Regimen C induction and those with slow morphologic early response or MRD positivity, SCT in CR1. However, the toxicity of SCT combined with recent results of recent European protocols suggests that

many such patients may be cured without SCT in CR 1. Thus at present there is no strong reason to recommend SCT independent of treatment response in this group.

t(17;19)(q22;p13)/TCF3(E2A)-HLF

A variant of the t(1;19) (E2A-PBX), t(17;19) (E2A-HLF) is a rare cytogenetic abnormality associated with BCP phenotype, DIC, hypercalcemia and a very high risk of early refractory relapse. It can be detected by routine karyotyping and FISH, and cytogenetics laboratories have been notified by the LRCG to report it as a high risk abnormality. In ALL97 all four patients with this abnormality relapsed between 5 and 18 months from diagnosis and subsequently died. Hence UK ALL 2003 recommended that these children receive Regimen C followed by allogeneic transplant after recovery from consolidation using any available donor. Two such SCT were performed in UKALL 2003, both patients relapsing soon after SCT. Thus at present no curative therapy is known for this group and maximal intensity induction and consolidation followed by an MRD directed decision re SCT seems appropriate.

MLL (KMT2A) rearrangement

Up to 2% of non-infant ALL are characterised by MLL (KMT2A) rearrangements confirmed on FISH. Analysis of ALL 97 reveals two subgroups of MLL (KMT2A) rearrangements- those with t(4;11) (q21;q23), who had a relatively good prognosis and those with other MLL (KMT2A) rearrangements who had a poorer outcome. In ALL 99 and 2003 all patients with MLL rearranged ALL were treated with regimen C and those with M2 marrow at day 28 were allocated SCT. With this approach only 1 of 19 patients with t(4;11) rearrangement have relapsed whereas 6 of the 26 with other MLL rearrangements have suffered a relapse. Given that at present there is no good evidence of the value of SCT in MLL rearranged ALL in CR1 maximal intensity induction and consolidation followed by an MRD directed decision regarding SCT seems appropriate.

Near haploidy (<30 chromosomes) and Low Hypodiploidy (30-39 chromosomes)

Up to 5% of ALL are characterised by the presence of less than 46 chromosomes or hypodiploidy. Several groups, including CCG, have reported a poor prognosis for hypodiploid disease. Further analysis of ALL97 revealed three distinct subgroups of hypodiploidy. High hypodiploid ALL (40-45 chromosomes) had a relatively good prognosis, whereas for those with low hypodiploid ALL (30-39 chromosomes) and near haploid ALL (<30 chromosomes) had a significantly poorer outcome. On the basis of these findings in ALL99 and ALL2003 all patients with hypodiploid ALL characterised by less than 44 chromosomes were treated with regimen C. Those with M2 marrow at day 28 were allocated SCT. With this approach the 5 year EFS of high hypodiploid disease on ALL 99 was 87% whereas that of low hypodiploid and near haploid disease 50%. At present there is no good evidence of the value of SCT in CR1 for low hypodiploidy and near haploid disease and thus maximal intensity induction and consolidation followed by an MRD directed decision re SCT seems appropriate. The definition of high risk hypodiploidy as <40 chromosomes is based on further analysis of UKALL 2003. Specifically, examination of UKALL 2003 samples with hypodiploid (40-43 chromosomes) karyotypes revealed extensive clonal heterogeneity making a clear distinction between cases with 40-43 and 44-45 chromosomes very difficult. In addition, patients with 40-43 chromosomes were very rare and did not have a poor outcome.

1.2.8 Definition of good risk cytogenetic abnormalities in B-cell precursor ALL

Section 1.2.1 outlines the rationale for using a different MRD threshold for patients with good and intermediate risk cytogenetics. For the purposes of this trial the following two chromosomal abnormalities will be classified as good risk. High risk chromosomal abnormalities are defined in section 1.2.7. All other patients, including those with t(1;19)(q23;p13) and B-other ALL are classified as having intermediate risk cytogenetics; including those with a missing or failed genetic analysis.

ETV6-RUNX1 (formerly known as TEL-AML1)

This gene fusion arises from the t(12;21)(p13;q22) translocation and is present in ~25% paediatric patients with B-cell precursor ALL. This gene fusion is cytogenetically cryptic and therefore detection must be determined by FISH using commercially available FISH probe, for example the dual colour dual fusion ETV6-RUNX1 translocation probes or RT-PCR using established primers.

High hyperdiploidy (51-65 chromosomes)

High hyperdiploidy is readily detectable by cytogenetic analysis and is present in upto one third of paediatric patients with ALL. The gain of between 5 and 19 extra chromosomes takes the modal chromosome number to between 51 and 65 inclusive. Detection of high hyperdiploidy by FISH requires the use of multiple probes which must include centromeric FISH probes to some of most frequently gained chromosomes (X, 4, 6, 10, 14, 17, 18, 21). In addition to evidence for the presence of at least 5 extra chromosomes, there must also be evidence for the absence of triploidy and tetraploidy (the gain of 1 or 2 whole extra sets of chromosomes). This can be achieved by using centromeric FISH probes for rarely gained chromosome (3, 7, 11, 16 and 19).

High hyperdiploidy should NOT be diagnosed by DNA index or solely via the use locus-specific FISH probes.

Please note that rare patients with t(9;22)/BCR-ABL1, t(1;19)TCF3-PBX1, iAMP21, MLL (KMT2A) gene fusions or T-ALL can have karyotypes with 51-65 chromosomes. Here the high hyperdiploidy is a secondary feature and they should not be classified as having good risk cytogenetics. In addition although patients with near-haploidy (23-29 chromosomes) is very rare, they can present with chromosomal doubling and an abnormal karyotype comprised entirely (or mostly) of tetrasomies and a modal chromosome number 46-58. These patients should not be confused with high hyperdiploidy.

2. AIMS, OBJECTIVES AND OUTCOME MEASURES**2.1 Aims and Objectives****2.1.1 Aims**

To define whether further refinement of MRD based risk stratification and treatment regimen improves survival whilst reducing overall burden of therapy in children and young adults suffering from ALL or LBL (T-cell non-Hodgkin's Lymphoma (NHL) or Smlg-ve precursor B-NHL).

2.1.2 Objectives

Randomised

1. To reduce toxicity through introduction of a short 14-day course of high dose dexamethasone in lieu of the conventional lower dose given for 28 days in induction. Randomisation closed 31st March 2017.
2. To provide more effective CNS prophylaxis and reduce burden of therapy through introduction of high dose methotrexate and by omission of vincristine and dexamethasone pulses and continuing intrathecal therapy in maintenance

Non – randomised

3. To decrease toxicity and reduce burden of therapy by administering a single delayed intensification to all patients and limiting augmented therapy to those who are not MRD Low Risk.

2.2 Outcome Measures

2.2.1 Primary outcome measures

4) Dexamethasone Randomisation (1st Randomisation, R1) (refer to Trial Design for update on closure of R1)

1. Induction steroid-induced morbidity and mortality defined as all serious, grade 3 or 4 adverse events related to induction and categorised as steroid related or steroid contributory and all treatment related deaths during induction.

2. Methotrexate Randomisation (2nd Randomisation, R2)

Central nervous system (CNS) relapse, defined as any relapse with CNS involvement, including combined.

3. Pulses Randomisation (2nd Randomisation, R2)

Bone marrow relapse, defined as any relapse with bone marrow involvement, including combined, Quality of Life measured by PedsQL.

Any event defined as relapse, secondary tumour or death from any cause is also a primary outcome measure for each randomised comparison and the trial overall.

2.2.2 Secondary outcome measures

1. Dexamethasone Randomisation (R1)

Rate of remission, event free and overall survival.

2. Methotrexate Randomisation (R2)

Event free and overall survival, Quality of Life measured by PedsQL, treatment related mortality and morbidity

3. Pulses Randomisation (R2)

Event free and overall survival, treatment related mortality and morbidity, local relapse (LBL)

4. Event free and overall survival, relapse rate, treatment related mortality and morbidity compared to the results of the UKALL 2003 trial.

R2 is a factorial randomisation

Clinical outcomes will be measured using internationally accepted definitions of relapse and treatment-related mortality (TRM), and NCI CTCAE toxicity grades (see Appendix 3 for CTCAE version 4.0). For the dexamethasone randomisation, steroid induced morbidity in induction will be assessed as any induction treatment related death and all serious adverse events (SAEs), within 8 weeks of the start of induction, which are classified as steroid related or contributory. I.e. events reported on the Steroid AE form or classed as possibly, probably or definitely related to dexamethasone given in induction on an SAE form. Steroid related AE refers to any events known to be directly caused by exposure to high dose glucocorticoids namely diabetes, psychosis, hypertension, obesity and pathological osteopenia or fracture. Steroid contributory refers to those in which exposure to steroid is a significant contributory factor namely severe bacterial, fungal or viral infection, pancreatitis, thrombosis and encephalopathy. Quality of Life (QoL) will be assessed using questionnaires, originally designed by Varni, to score generic and disease specific measures in a paediatric population validated in UKALL 2003 (see Appendix 13).

3. TRIAL DESIGN

The UKALL2011 trial is a multicentre, phase III, randomised controlled trial. The trial is designed to improve survival and quality of survival through further refinement of the Children's Cancer Group (CCG) treatment regimens (Regimens A, B and C) used in UKALL 2003. As in UKALL 2003 all patients receive dexamethasone and pegaspargase (Oncaspar, pegylated e-coli asparaginase). Pegaspargase is used throughout. Greater emphasis is placed on toxicity and quality of life as trial end points in UKALL 2011 than was done in UKALL 2003 and collection of this data is fundamental to the success of the trial.

3.1 Changes to treatment regimens compared to UKALL 2003

UKALL 2011 carries forward the regimens A, B and C employed in UKALL 2003. In addition to the changes in therapy to be explored by randomisation attention is drawn to the following changes to the regimens used in UKALL 2003.

- Induction therapy is **not** modified according to day 8 or 15 response (apart from NCI High Risk Down syndrome patients where day 15 marrow may alter induction treatment – see Appendix 5 for details).
- All patients with T-ALL receive a four drug induction (Regimen B Induction).
- Five high risk cytogenetic abnormalities are recognised in UKALL 2011: iAMP21, t(17;19)(q22;p13), MLL (KMT2A) rearrangements, near haploidy and low hypodiploidy.
- All patients receive a single delayed intensification.
- An additional bone marrow sample is required at week 9 of Regimen C (ALL patients only). This will be used in a prospective blinded analysis of MRD clearance in order to improve understanding of MRD kinetics in this group.
- Introduction of an additional week of low dose mercaptopurine at the end of consolidation in Regimen C.
- Standard interim maintenance now lasts 9 weeks in regimens A and B.
- No intrathecal therapy is given in the second part of DI in regimens A and B.
- No bone marrow aspirates are required after day 29 in MRD Low Risk and after recovery from consolidation (week 14) in MRD Intermediate Risk patients.
- An additional intrathecal dose in standard interim maintenance in Regimen A.

3.2 Stratification of treatment in UKALL 2011

3.2.1 Acute Lymphoblastic Leukaemia

Four non-randomised modifications to the stratification used in UKALL 2003 are employed in UKALL 2011.

- 1) All patients with T-cell ALL receive a four drug induction (Regimen B induction).
- 2) Post-induction therapy is allocated on the basis of day 29 MRD in all patients regardless of early response. For MRD No Result patients, post-induction therapy is determined by early response as assessed by morphology and no further MRD measurement is required.
- 3) All patients receive a single DI.
- 4) Patients with persistent high level MRD (MRD High Risk) after augmented BFM consolidation in Regimen C are taken off protocol treatment and allocated SCT in First Complete Remission (CR1).

Thus in UKALL 2011 treatment intensity is stratified as follows:

- In precursor B-ALL induction treatment is allocated according to NCI Risk. NCI Standard Risk patients (Age >1 year and <10 years and with a highest WCC before starting treatment of $<50 \times 10^9/L$) receive three induction drugs (Regimen A induction). NCI High Risk patients (Age ≥ 10 years and/or with a diagnostic WCC $\geq 50 \times 10^9/L$) receive four induction drugs (Regimen B induction).

- All patients with T-cell ALL are categorised as NCI High Risk and receive four induction drugs (Regimen B induction).
- Stratification by early morphologic response will be abandoned in all patients with an adequate day 29 MRD result (estimated to be 93% of entrants). Patients with no MRD result at day 29 due to an inadequate sample or no MRD marker are assessed by morphology. Patients with a slow early response (SER) receive Regimen C treatment following induction therapy. Those with a rapid early response (RER) receive Regimen A or Regimen B following induction as per the treatment arm assigned for induction.
- Post-induction therapy will be stratified by cytogenetic risk and MRD level at day 29 and week 14 (see section 1.2.8 for definition of cytogenetic risk groups).
- Those with MRD $<0.005\%$ (cytogenetic intermediate risk); or $<0.1\%$ (cytogenetic good risk) at day 29 (at least one marker quantitative range of 10^{-4}) form the **MRD Low Risk group**. MRD Low Risk BCP ALL receive standard NCI directed post-induction consolidation therapy (Regimen A or B) and a single DI. MRD Low Risk T-ALL receive standard Regimen B post-induction therapy. No further MRD measurement is undertaken in this group.
- Patients with MRD $\geq 0.005\%$ (cytogenetic intermediate risk); or $\geq 0.1\%$ (cytogenetic good risk) at day 29 form the **MRD Risk group** and receive augmented BFM consolidation therapy (Regimen C consolidation).
- In the **MRD Risk group**, MRD is reassessed upon count recovery from consolidation. Those with levels $\geq 0.5\%$ are deemed **MRD High Risk** and taken off protocol treatment (allocation of SCT in the national relapse refractory study should be considered). Those with levels $<0.5\%$ are deemed **MRD Intermediate Risk** and continue Regimen C with a single DI.
- Both the MRD Low Risk and Intermediate Risk groups are eligible for the methotrexate and pulses randomisation (R2).

3.2.2 Lymphoblastic Lymphoma

In line with current international practice all patients with LBL receive a four drug induction (NCI High Risk, Regimen B induction).

Following induction, further therapy is determined according to tumour response:

Patients with $<35\%$ tumour volume reduction after induction therapy are considered poor responders and are taken off UKALL 2011 protocol treatment. Their care should be discussed with either the Chief Investigator or the LBL coordinators.

Patients with $\geq 35\%$ tumour volume reduction are deemed to be in remission and if of T immunophenotype receive augmented BFM consolidation (Regimen C consolidation), and continue therapy as per Regimen C with a single DI. These patients are eligible for the factorial methotrexate and pulses randomisation (R2) within Regimen C. Patients of precursor B immunophenotype receive standard Regimen B post induction therapy. These patients are eligible for the factorial methotrexate and pulses randomisation (R2) within Regimen B.

3.3 Allocation of induction therapy

All patients registered on the study are eligible to participate in the cell banking, cytogenetic, Quality of life and other add on biological studies, with appropriate consent, maximising the population based knowledge of leukaemia biology.

A minimal dataset will be collected on patients registered after appropriate consent – see section 9.1.

All patients who consent to enter the trial must be registered within 7 days of starting induction treatment.

Induction therapy is stratified as follows **BCP ALL**:

Patients with B-Cell precursor ALL (BCP ALL) should receive NCI risk-assessed induction.

NCI Standard Risk: Patients aged ≥ 1 year and < 10 years old at diagnosis and with a highest white blood cell count (WCC) before starting treatment of $< 50 \times 10^9/L$.

Patients in this group receive a 3-drug (dexamethasone, vincristine and asparaginase) induction (**Regimen A induction**).

NCI High Risk: Patients aged ≥ 10 years old at diagnosis, and/or with a diagnostic WCC $\geq 50 \times 10^9/L$.

Patients in this group received a 4-drug (dexamethasone, vincristine, asparaginase and daunorubicin) induction (**Regimen B Induction**).

T-CELL ALL AND LBL PATIENTS

All patients with T cell ALL (independent of NCI risk) or LBL should receive induction treatment as per the NCI High Risk induction regimen (**Regimen B Induction**).

Please Note

In UKALL 2011 induction is NOT modified according to day 8 or 15 morphology, however Down syndrome patients with SER at day 15 may be intensified – see section 3.3.2.

3.3.1 Modification of Induction for High Risk Cytogenetics

A small number of NCI Standard Risk BCP ALL patients initially treated according to Regimen A induction will subsequently be found to have high risk cytogenetics (MLL (KMT2A) rearrangements, near haploidy, low hypodiploidy, iAMP21, and t(17;19)) and should transfer to Regimen C induction on day 15. These patients will then receive Regimen C consolidation and further therapy will be guided by MRD upon recovery from Regimen C consolidation.

Any patient with T-ALL, B-LBL or NCI High Risk BCP ALL that also has high-risk cytogenetics (MLL (KMT2A) rearrangements, near haploidy, low hypodiploidy, iAMP21, and t(17;19)) should be given Regimen B induction and transfer to Regimen C at the beginning of consolidation (see Figures 2 and 4). Their further therapy will be guided by MRD upon recovery from Regimen C consolidation.

3.3.2 Induction for Down Syndrome patients (ALL and LBL)

Induction for Down Syndrome patients (ALL and LBL) In line with international experience, Down syndrome (DS) patients treated on ALL 99 and UKALL 2003 experienced excess treatment related death. Consequently in UKALL 2011 these patients receive a modified protocol (see Appendix 5) Thus, patients with Down syndrome should NOT initially receive anthracycline in induction i.e all DS patients should receive Regimen A induction.

However, Down syndrome patients with T-ALL and NCI High Risk BCP ALL and a slow early response at day 15 or slow resolution of bulk disease in LBL, s should switch to Regimen C induction at day 15, if there is no serious morbidity to preclude doing so. See Appendix 5 for further details.

Down syndrome patients are not eligible for the methotrexate and pulses randomisation (R2). See Appendix 5 for further details on the treatment of DS patients.

3.4 Allocation of post-induction therapy in patients with ALL

3.4.1 Obtaining a Day 29 MRD result

All patients with ALL have a bone marrow test for Minimal Residual Disease (MRD) at day 29 of induction. A MRD sample should be sent to the MRD laboratory as per the guidelines in Section 7.9 and Appendix 4.

N.B. Day 29 marrow should be delayed if neutrophils $<0.5 \times 10^9/L$ or platelets $<50 \times 10^9/L$.

3.4.2 Allocation of post-induction therapy (Day 29 MRD Result)

Post-induction therapy is stratified according to the day 29 MRD result as described below.

MRD Low Risk

MRD Low Risk patients are defined as those with MRD $<0.005\%$ (**cytogenetic intermediate risk**); or **$<0.1\%$ (cytogenetic good risk)** at day 29 using at least one marker with a quantitative range of 10^{-4} . These patients should receive consolidation as per the treatment arm previously assigned for induction (**Regimen A or B**). No further MRD measurement is required. These patients are eligible for the methotrexate and pulses randomisation (R2).

Randomisation should take place **as soon as possible after obtaining the day 29 MRD result** and after informed consent for the second randomisation (R2) has been obtained.

MRD Risk

MRD Risk patients are defined as those with evidence of MRD at a level $\geq 0.005\%$ (**cytogenetic intermediate risk**) or **$\geq 0.1\%$ (cytogenetic good risk)** at day 29 using at least one marker with a quantitative range of 10^{-4} . These patients should receive augmented BFM consolidation (**Regimen C consolidation**) and have further MRD assessment upon count recovery from consolidation at week 14.

MRD No Result.

MRD No Result patients are defined as those with no MRD result at day 29 due to inadequate samples or no MRD marker with a quantitative range of 10^{-4} . Experience in UKALL 2003 shows that a MRD result (one marker with a quantitative range 10^{-4}) can be provided for 93% of patients in whom adequate samples are received.

Post-induction consolidation therapy in these patients is determined by early response as assessed by morphology:

- Patients with a **slow early response (SER)**, defined as $\geq 25\%$ blasts at day 8 of induction as per Regimen B or day 15 of induction of Regimen A receive **Regimen C consolidation**.

These patients then continue therapy as per **Regimen C**. No further MRD assessment is required in these patients.

- Patients with a **rapid early response (RER)**, defined as <25% blasts at day 8 of induction as per Regimen B or day 15 of induction of Regimen A continue therapy as per **Regimen A or B as per treatment arm previously assigned** for induction. No further MRD assessment is required in these patients.

All MRD No Result patients are eligible for the methotrexate and pulses randomisation.

For high risk cytogenetics patients who are MRD No Result, please discuss further with the Chief Investigator or Co-investigators.

Figure 2: Allocation of post-induction therapy in patients with ALL

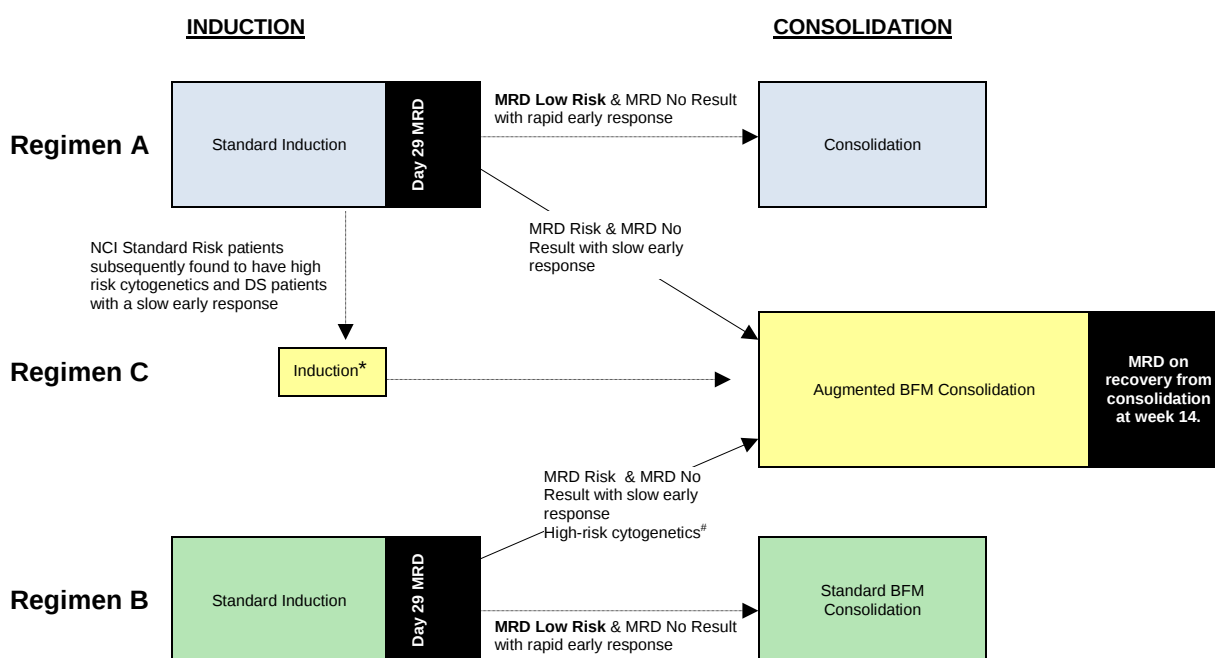


Figure 2

Allocation of post-induction therapy for patients with ALL. Post-induction therapy is stratified according to the day 29 MRD result. For MRD No Result patients, those with a slow early response receive Regimen C consolidation. No further MRD assessment is required in these patients and they continue Regimen C. MRD No Result patient with a rapid early response continue therapy as per Regimen A or B as previously assigned.

*Patients with high risk cytogenetics initially started on Regimen A induction transfer to Regimen C induction on day 15. Also Down syndrome patients who have a slow early response on day 15 bone marrow should transfer to regimen C induction on day 15.

[#] Patients known to have high risk cytogenetics at the start of treatment and treated with Regimen B induction transfer to Regimen C consolidation.

3.4.3 Obtaining a post consolidation MRD result (MRD Risk group only)

For MRD Risk patients, a MRD sample should be obtained upon count recovery from Regimen C consolidation at week 14 and sent to the relevant MRD laboratory as per the guidelines in section 7.9 and Appendix 4. Once the sample has been taken consideration should be given to starting the low dose holding chemotherapy as per week 15 of Regimen C consolidation. The laboratory will notify you of the patient's MRD result within 5 working days of receipt of the sample. Please note that patients in the MRD Low Risk group at day 29 do not have a post-consolidation MRD measurement.

N.B. The post consolidation marrow should be delayed if neutrophils <0.5X10⁹/L or platelets <50X10⁹/L.

3.4.4 Allocation of post consolidation therapy (MRD Risk group only)

In the MRD Risk group further therapy is stratified according to the post-consolidation MRD result as described below (see also Figure 3). MRD Risk patients are categorised as 'MRD Intermediate Risk' or 'MRD High Risk' based on the results of the post consolidation MRD result.

MRD Intermediate Risk

MRD Intermediate Risk patients are defined as those with no evidence of MRD at a level of or greater than 0.5% (MRD $<0.5\%$) after Regimen C augmented BFM consolidation using at least one marker with a quantitative range of 10^{-4} . These patients **continue therapy as per Regimen C** and are eligible for the methotrexate and pulses randomisation (R2).

Randomisation should take place as soon as possible after receiving the post-consolidation MRD result and informed consent for the second randomisation (R2) has been obtained.

MRD High Risk

MRD High Risk patients are defined as those with evidence of MRD at a level $\geq 0.5\%$ after Regimen C augmented BFM consolidation. These patients are at high risk of relapse and are taken off UKALL 2011 protocol treatment. It is recommended that these patients be screened for the presence of a targetable tyrosine kinase alteration (see 3.7.5.4). Further therapy for these patients should be discussed with the Chief Investigator or Co-Investigators and will likely include novel cytoreduction prior to SCT

Figure 3: Allocation of post-consolidation therapy for MRD Risk patients

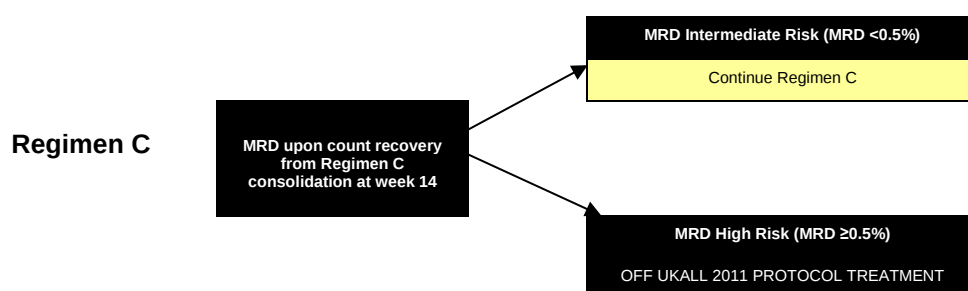


Figure 3

MRD is re-assessed upon recovery from consolidation at week 14 for MRD Risk patients (Regimen C). Patients with $\geq 0.5\%$ MRD are taken off UKALL 2011 protocol treatment. Patients with $<0.5\%$ MRD continue treatment on Regimen C.

3.5 Allocation of post-induction therapy in LBL patients

Volumetric measurement of tumour mass should be made as soon after presentation as it is safe to do so. For T-NHL, assessment by CT scan is recommended. The same modality of imaging should then be used to calculate the reduction in tumour volume at day 29. It is important to recognise that tumour regression is frequently incomplete. PET scanning is unproven in LBL and not currently recommended for stratification of treatment.

Post-induction therapy is stratified according to the day 29 assessment result as described below (see also Figure 4)

Patients with <35% reduction in tumour volume are considered poor responders and are taken off protocol treatment. Their further therapy should be discussed with the Chief Investigator or Co-investigators.

Patients with $\geq 35\%$ reduction in tumour volume are considered good responders and those of T immunophenotype receive Regimen C and are eligible for the methotrexate and pulses randomisation (R2). Those patients with precursor B immunophenotype LBL continue Regimen B and are eligible for the methotrexate and pulses randomisation (R2).

Figure 4: Allocation of post-induction therapy for LBL patients

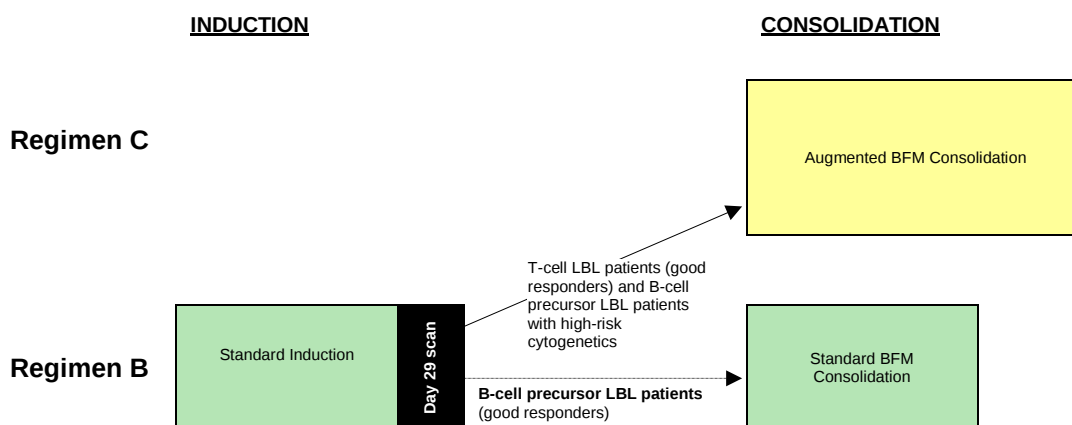


Figure 4

Allocation of post- induction therapy for patients with LBL. Post-induction therapy is stratified according to the immunophenotype and day 29 scan result.. T-cell LBL patients with a good response receive Regimen C consolidation and continue Regimen C thereafter. Precursor B-cell LBL patients continue Regimen B throughout, unless they have high-risk cytogenetics. Precursor B-cell LBL patient with high-risk cytogenetics transfer to Regimen C at the end of induction.

3.6 Methotrexate and Pulses Randomisation (R2)

This randomisation is only open to LBL good responders and those patients with ALL in the MRD Low Risk, MRD Intermediate Risk, and MRD No Result groups.

Down syndrome patients are not eligible for this randomisation.

In MRD Low Risk and MRD No Result patients consent should be obtained for the methotrexate and pulses randomisation as soon as possible after receipt of the day 29 MRD result.

In LBL good responders, consent should be obtained for the methotrexate and pulses randomisation (R2) as soon as possible after receipt of the day 29 tumour volume assessment.

In MRD Intermediate Risk patients, consent should be obtained for the methotrexate and pulses randomisation (R2) as soon as possible after receipt of the day 29 MRD result. However, the patient should not be randomised until the receipt of the MRD result taken upon recovery from consolidation at week 14 (ALL patients).

See Figure 5 for a summary of the different treatment regimens resulting from R2.

3.7 Special Categories

3.7.1 CNS Disease at Presentation (CNS3):

Acute lymphoblastic leukaemia

No patient with ALL will receive cranial irradiation in UKALL 2011. It is recommended that patients with CNS disease at diagnosis (CNS3 status - the presence of $>5/\text{mm}^3$ unequivocal lymphoblasts in the CSF) have the diagnosis verified by central review of CSF cytology (slides should be sent to the Chief Investigator within one month of diagnosis). These patients will receive weekly intrathecal methotrexate for a total of 4 intrathecal during weeks 1-4 of induction.

Patients in whom the CSF is clear by day 29 will continue NCI and MRD directed therapy. These patients are eligible for R2.

Patients with persisting CNS disease at the end of induction will transfer to Regimen C at the end of induction, regardless of bone marrow response. If they clear CNS disease subsequently and are $<0.5\%$ MRD Intermediate Risk at week 14, they remain eligible for R2. Those with MRD $\geq 0.5\%$ are deemed MRD High Risk and are not eligible for R2. They should be discussed with the Chief Investigator or Co-investigators.

MRD No Result patients with persistent CNS disease at the end of induction should be discussed with the Chief Investigator or Co-investigators.

Lymphoblastic Lymphoma

In LBL patients with CNS disease at diagnosis (CNS3 status - the presence of $>5/\text{mm}^3$ unequivocal lymphoblasts in the CSF) it is recommended that diagnosis is verified by central review of CSF cytology (slides should be sent to the Chief Investigator). They will receive weekly intrathecal methotrexate for a total of 4 intrathecal during weeks 1-4 of induction. **Patients in whom the CSF is not clear by day 29 should be discussed with the Chief Investigator or LBL Co-investigators.**

3.7.2 Traumatic or CNS2 LP at diagnosis

There is evidence that a traumatic lumbar puncture (LP) with blasts (TLP+) or CNS2 (atraumatic with <5 blasts/microlitre) tap at diagnosis is associated with a higher risk of CNS relapse, possibly because of poor penetration of MTX into the meningeal space at subsequent treatments due to a small extradural haematoma at the LP site. Therefore, patients with >10 red cells/ μL and blasts in their CSF or CNS2 tap at diagnosis should receive weekly intrathecal MTX for a total of 4 intrathecal during weeks 1-4 of induction. Traumatic tap without blasts in the CSF should receive standard intrathecal therapy. These patients are eligible for both randomisations.

Please note that the Chief Investigator and Co-investigators strongly advise against performing diagnostic lumbar punctures in ALL without introduction of intrathecal chemotherapy at diagnosis.

3.7.3 Testicular Disease at Diagnosis

Patients with clinically enlarged testis at presentation should be assumed to have testicular disease.. Biopsy is not required to confirm the presence of infiltrate. Routine ultrasound of the testis should not be performed to detect sub-clinical enlargement.

These patients are eligible for both registration and R2. Patients with testicular disease should start induction according to NCI risk (age and WCC count) and disease type. Post-induction therapy should be stratified by MRD. Standard treatment for those patients who still have a clinically enlarged testis at week 8 is testicular radiotherapy. Testicular radiotherapy should be given during the first cycle of maintenance - guidelines can be found in Appendix 10.

3.7.4 High risk cytogenetics

As described in section 1.2.7 five high risk cytogenetic abnormalities are recognised in UKALL 2011: iAMP21, t(17;19), MLL (KMT2A) rearrangements, near haploidy and low hypodiploidy.

Patients known to have high risk cytogenetics at the start of treatment should receive a four drug induction (Regimen B) followed by Regimen C augmented BFM consolidation. Further therapy will be directed by the MRD result upon recovery from consolidation at week 14.

A small number of NCI Standard Risk BCP ALL patients initially treated according to Regimen A induction will subsequently be found to have high risk cytogenetics and transfer to Regimen C induction at day 15 of induction. These patients then receive Regimen C consolidation and further therapy is directed by the MRD result upon recovery from consolidation.

NB: Patients with intrachromosomal amplification of chromosome 21 (iAMP21)

Please note that not all patients with multiple copies of RUNX1 by FISH fit into this category. Please discuss the result with your cytogenetic laboratory and, if in any doubt, contact Professor Christine Harrison and/or Professor Anthony Moorman at the Cytogenetics Coordinating Centre (contact details can be found in Appendix 20).

NB: Patients with MLL (KMT2A) rearrangements

Please note that these recommendations only apply to patients with a confirmed MLL translocation – either by FISH or the presence of an established MLL (KMT2A) /11q23 translocation by cytogenetics. Patients with MLL (KMT2A) deletions should receive standard NCI risk directed therapy. If uncertain please discuss result with your cytogenetics laboratory and, if in any doubt, contact Professor Christine Harrison and/or Professor Anthony Moorman at the Cytogenetics Coordinating Centre (contact details can be found in Appendix 20).

3.7.5 Induction Failure: defined as M3 marrow and/or molecular MRD >5%. In the absence of molecular MRD, M2 marrow confirmed on central review by CI or co-investigators.

3.7.5.1 BMA M3 (≥ 25% blast at day 29) or T-ALL with BM1 and MRD >10%

Unequivocal presence of ≥ 25% blasts at day 29, was rare (<1% of all cases) in ALL 99 and 2003. There is data both from the BFM and an international collaborative study that such patients benefit from SCT in CR1. However even with SCT there is a high risk of relapse and such patients may benefit from immune therapy for B-lineage and Nelarabine for T-lineage disease. Thus in UKALL 2011, patients with an M3 marrow at day 29 will come off protocol treatment. Investigators may wish to discuss alternative therapy with the Chief Investigator or Co-investigators. Slides from diagnosis and day 29 should be sent to the Chief Investigator for review.

All patients with M3 marrow should be screened for a targetable tyrosine kinase alteration (see 3.7.5.4) and appendix 25

During the first years of UKALL 2003 it became apparent that patients with T-ALL and a molecular MRD level of > 10% had an extremely poor prognosis (7 patients. 6 early marrow relapses) despite being in morphological remission. From that point on, it was recommended that these patients should be treated with a regimen based on Nelarabine and consolidate with SCT. Although there has been no formal analysis of outcome of this strategy anecdotal UK evidence that this approach improves prognosis.

3.7.5.2 MRD ≥5% BMA M2 confirmed by central review

In UKALL 2003 29 patients had a M2 marrow at day 28, all of whom were transferred to arm C. Subsequently 25 achieved a remission and 4 died with refractory disease. Of the 25 who remitted 5 had SCT of whom 2 relapsed, whereas 20 received chemotherapy all of whom remain in remission with a median follow-up of 2.5 years. Six patient with M2 marrows had MRD <0.01%, all of whom survived.

Recent review of D29 MRD level and outcome on UKALL2003 showed an additional 48 patients

(1.8%) with M1 marrow and MRD >5% who had an EFS of 43.9% and an overall survival of 63.8%, near identical to those with M2 marrow (EFS 57.1%, OS 61.1%). However, the outcome of NCI standard risk patients and those with good risk cytogenetics was better, especially if they received Regimen C post-remission.

Thus, these patients should receive Regimen C consolidation followed by treatment depending on NCI risk, genetic risk and response at week 9/14. All such patients should also be screened for ABL kinase fusion abnormalities (see 3.7.5.4 and appendix 25) and discussed with the Chief Investigator or Co-investigators.

Clinicians will be informed by the MRD laboratories if a patient has induction failure on the basis of MRD.

3.7.5.3 B-ALL with slow MRD response or induction failure and ABL-class fusions – see appendix 25

3.7.5.4 Targetable tyrosine kinase alterations (see appendix 25)

Recent molecular genetic research has revealed a series of novel translocations and rearrangements which result in tyrosine kinase activation and sensitivity to a tyrosine kinase inhibitor (TKI) (Cancer Cell 2012 v22, p153; ASH Educ Program. 2013, p118). These rearrangements involve the fusion of a kinase gene (e.g. *ABL1*, *ABL2*, *CSF1R* or *PDGFRB*) to wide variety of “activating genes” (e.g. *PAX5*, *EBF1*, *NUP214*, *ZMIZ1*, etc). A recent study in the New England Journal of Medicine (NEJM) (2014 v371, p1005) has extended our knowledge of this network of gene fusions.

From our own FISH screening studies (unpublished) and the NEJM study, we estimate that the frequency of these TKI-sensitive gene fusions is ~1% in standard risk childhood ALL and 2-3% in high risk childhood/adolescent ALL. Although most of these cases will respond well to conventional MRD based therapy, there is good evidence (albeit from <20 patients) that patients with one of these gene fusions who have slow MRD clearance will respond favourably to imatinib. Thus it is recommended that all patients with MRD >1% or M2 or M3 at the end of induction or MRD > 0.5% at week 14 marrow are screened for these lesions. Please note that routine screening of the MRD low and intermediate risk groups is not recommended.

A full guideline on screening for targetable tyrosine kinase alterations is provided in appendix 25. Please read this guideline in full prior to contacting the local genetics laboratory or the coordinating genetic laboratory to request additional testing.

3.8 Duration of treatment

For all patients (ALL and LBL) treatment will last for exactly 2 years from the start of interim maintenance for female patients and 3 years from the start of interim maintenance for male patients. The cycle in progress is stopped when this date is reached.

3.9 Frequency and duration of follow-up

During (registration treatment) Induction and consolidation treatment a minimal dataset will be recorded (see section 9.0) using case reports forms (CRFs).

For participants subsequently randomised to R2, clinical outcomes will be measured during, interim maintenance and delayed intensification and after each cycle of maintenance treatment. After completion of therapy, remission status will be ascertained by annual questionnaire to treatment centres for a minimum of five years following completion of maintenance therapy.

4. ELIGIBILITY

4.1 Inclusion Criteria

Registration to UKALL 2011 is open to all patients from age 1 (first birthday) to 24 years 364 days (at time of diagnosis) with a first diagnosis of acute lymphoblastic leukaemia or lymphoblastic lymphoma (T-NHL or Smlg negative precursor B-NHL) diagnosed using standard criteria. Written informed consent is required for all patients and a negative pregnancy test within 2 weeks prior to starting treatment for female patients of childbearing potential. Exclusion Criteria

4.1.1 Trial entry

The following patients are excluded from entering the trial

1. Infants less than a year old **at diagnosis**. It is recommended that these patients be entered onto the relevant Interfant ALL study.
2. Patients diagnosed with mature B-ALL (Burkitt-like, t(8;14), or C-MYC re-arranged regardless of morphology or phenotype)). Patients with this disease should be treated as mature-B-ALL on a suitable protocol for this condition.
3. Patients diagnosed with Philadelphia-positive ALL (t(9;22) or BCR/ABL positive). If randomised/registered patients are subsequently found to have Philadelphia-positive ALL they will be withdrawn from UKALL 2011 protocol treatment and transferred to a suitable alternative protocol for further therapy.
4. Patients in whom written informed consent has not been obtained from parents and/or patients prior to trial entry
5. Patients who are sexually active and are unwilling to use adequate contraception during therapy and for one month after last trial treatment.

To be considered for R2, patients must have been registered to UKALL 2011.

4.1.2 Exclusion criteria for R2 (methotrexate and pulses randomisation).

1. MRD High Risk ALL patients as defined in section 3.4.4 and LBL patients with a poor response as defined in section 3.5. These patients are taken off protocol treatment.
2. B-lineage ALL patients with MRD >1% at day 29 of induction and ABL class fusion abnormalities. See appendix 25.
3. Any patient with significant renal impairment defined as renal function outwith normal limits corrected for age, pleural effusions or ascites. It is recommended that these patients receive standard A or B (MRD Low Risk) or Capizzi (Regimen C) interim maintenance (MRD Risk or LBL); vincristine and dexamethasone pulses and intrathecal methotrexate in maintenance.
4. Previous history of methotrexate encephalopathy. It is recommended that these patients receive standard (MRD Low Risk) or Capizzi (Regimen C) interim maintenance (MRD Risk or LBL) and vincristine and dexamethasone pulses during continuation therapy.
5. MRD Intermediate Risk patients with a history of pancreatitis. These patients should not receive further asparaginase. It is recommended that these patients be allocated high dose methotrexate as interim maintenance (without asparaginase) and vincristine and dexamethasone pulses during continuation therapy.
6. Candidates for allogeneic SCT in CR 1.
7. Down syndrome (DS) patients. See section 3.3.2 and Appendix 5 for further information about DS patients.
8. Patients who have received prior cranial irradiation.
9. Patients with M3 marrow at day 29.

5. SCREENING AND CONSENT

5.1 Screening

All patients with ALL and LBL from age 1 year (first birthday) to 24 years 364 days at first diagnosis will be screened for entry into this trial.

The following assessments and procedures should have been performed prior to trial entry.

- Full medical history and physical examination
- Height*, weight and body surface area (BSA)
- Assessment of performance status (Lansky or ECOG)
- Full blood counts and biochemistry
- Trephine biopsy and/or bone marrow aspirate.
- Pregnancy test for all female patients of child bearing age. This must be performed within 2 weeks prior to starting treatment.
- Confirmation of disease diagnosis from bone marrow aspirate (or peripheral blood where there is high presenting WCC) using immunophenotyping/flow cytometry, or CT scan/ultrasound for LBL patients (NB. Murphy staging should be used for LBL patients)
- Cytogenetic and molecular cytogenetic genetic analysis.
 - o All genetic testing should be performed by the regional genetic laboratory. It is expected that the laboratory will undertake a combination of cytogenetic analysis, FISH tests and molecular tests to accurately detect:
 - o ETV6-RUNX1 and high hyperdiploidy which are classified as good risk abnormalities – see section 1.2.8
 - o MLL (KMT2A) rearrangements, BCR-ABL1, iAMP21, t(17;19), near haploidy and low hypodiploidy which are classified as high risk abnormalities – see section 1.2.7
 - o ABL-class fusion in B-lineage ALL patients with induction failure or MRD slow response – see Appendix 25 for details

Sites must notify their MRD laboratory of a patient's cytogenetic risk group by completing a Clinical Update Form for MRD laboratories as soon as the information comes available. A copy of this form must also be sent to the Trials Office who will forward to the Leukaemia Research Cytogenetics Group (LRCG).

**It may not be feasible to obtain height at diagnosis in an acutely ill child. In most children drug dosing is based on weight alone see 7.4.1 for details*

CYTOGENETIC AND MOLECULAR CYTOGENETIC REPORTS

Copies of all diagnostic and relapse cytogenetic, FISH and genetic reports, along with the Clinical Update Form for MRD Laboratories must be sent to the Leukaemia Research Cytogenetics Group (LRCG) (see Appendix 20 for address) as soon as possible. In addition, the LRCG requires left-over fixed cell suspension, DNA, cDNA, RNA or other material. This material will be used to undertake further clinical investigations and research (e.g. the genomics sub study outlined in Appendix 20) All these additional tests will be performed with the full knowledge of the Chief Investigator and clinical coordinators.

NOTE REGARDING LUMBAR PUNCTURE FOR CSF CYTOLOGY

Samples should be examined for evidence of CNS disease through assessment of cell count and cytospin. Definitions of CNS disease are given in section 3.7.1. **Please note that the Chief Investigator and Co-investigators strongly advise against performing diagnostic lumbar**

punctures without introduction of intrathecal chemotherapy at diagnosis. The first lumbar puncture should ideally be performed when the first dose of intrathecal methotrexate is due.

NB administration of intrathecal methotrexate at the time of obtaining diagnostic CSF does not exclude the patient from trial entry.

5.1.1 Collection of diagnostic samples from LBL patients at diagnosis

In the management of a patient presenting with a mediastinal mass then immediate patient safety takes priority over establishment of a histological diagnosis. This is discussed further in Appendix 22. Expert anaesthetic opinion will guide the timing of any diagnostic procedure. Prior cytoreductive therapy that makes anaesthesia safe might be necessary prior to any diagnostic attempt. Procedures that might allow a diagnosis to be made without the need for general anaesthesia might be considered.

When a diagnostic procedure is safely attempted samples should be taken for histopathology including immunophenotyping in the first instance and then for cytogenetic analysis and for tumour storage for future research studies (cell banking). Under the same anaesthetic staging investigations including bone marrow and lumbar puncture should be undertaken.

5.2 Informed Consent

It is the responsibility of the Principal Investigator (or delegate if this duty has been delegated to a suitably qualified individual) to obtain written informed consent for each patient prior to performing any trial related procedure. Consent must be obtained separately for both registration and randomisation (R2) in this trial. Patient and Parent Information Sheets (PIS) are provided prior to registration and randomisation to facilitate this process. Investigators must ensure that they adequately explain the aim, trial treatment, anticipated benefits and potential hazards of taking part in the trial to the patient and/or parent as appropriate. The Investigator should also stress that the patient is completely free to refuse to take part and if they do decide to take part they can withdraw from the trial at any time. The patient/parent should be given ample time to read each Information Sheet and to discuss their participation with others outside of the site research team. The patient/parent must be given an opportunity to ask questions which should be answered to their satisfaction. The right of the patient or parent to refuse to participate in the trial without giving a reason must be respected.

If the patient/parent expresses an interest in participating in the trial at diagnosis they should be asked to sign and date the latest version of the Registration Informed Consent Form

Patient and Parent Information Sheets (PIS) for registration only are provided to facilitate this process.

The Investigator must then sign and date the form. A copy of the Informed Consent Form should be given to the patient/parent, a copy should be filed in the hospital notes, and the original placed in the Investigator Site File (ISF). Once the patient is entered into the trial the patient's trial number should be entered on the Informed Consent Form maintained in the ISF. In addition, if the patient has given explicit consent a copy of the signed Informed Consent Form must be sent in the **post** to the Trials Office for review.

Prior to randomisation (R2), the PIS for R2 (methotrexate and pulses randomisation) should be provided to the patient/parent. Further information about the timing of consent can be found in section 5.2.1.

Details of the informed consent discussions should be recorded in the patient's medical notes, this should include date of, and information regarding, the initial discussion, the date consent was given, with the name of the trial and the version number of the Patient Information Sheet and Informed Consent Form. Throughout the trial the patient should have the opportunity to ask questions about the trial and any new information that may be relevant to the patient's continued participation should be shared with them in a timely manner. On occasion it may be necessary to re-consent the patient or

parent in which case the process above should be followed and the patient's right to withdraw from the trial respected.

Electronic copies of the Patient and Parent Information Sheet and Informed Consent Form are available from the Trials Office and should be printed or photocopied onto the headed paper of the local institution.

Details of all patients approached about the trial should be recorded on the Patient Screening/Enrolment Log and with the patient's prior consent their General Practitioner (GP) should also be informed that they are taking part in the trial. A GP Letter is provided electronically for this purpose.

5.2.1 Stages of consent

CHILDREN UNDER 16 YEARS: Written informed consent must be given by parents or those with legal responsibility for children under 16 years of age but children should be asked for their assent, if appropriate.

PATIENTS OVER 16 YEARS: Written informed consent must be obtained from all patients aged 16 years or over.

UKALL 2011 includes two stages of consent 1) registration at diagnosis : and 2) (R2) the factorial methotrexate and pulses randomisation which is open to the MRD Low Risk and MRD Intermediate Risk groups for ALL, and good responders for LBL. Experience in UKALL 2003, revealed difficulties with obtaining consent for post-induction randomisations at diagnosis, a time when the patient's day 29 MRD risk or post-induction tumour assessment is unknown, therefore a two-step approach has been taken to gaining consent for the randomisations in the UKALL 2011 trial.

Acute Lymphoblastic Leukaemia

1. **At diagnosis**, provide parent/patient with the first (Registration) information sheet. Obtain consent for trial entry and data collection. In addition this PIS will include details of any additional tests, procedures or samples for which informed consent is required but are not mandatory for entry into the UKALL 2011 trial e.g. asparaginase study.
2. Then in **MRD Low Risk** and **MRD No Result** patients, provide parent/patient with the second (R2) information sheet and obtain consent for the methotrexate and pulses randomisation (R2) **as soon as possible following receipt of the day 29 MRD result.**

For MRD **Intermediate Risk** patients, provide parent/patient with the second information sheet and obtain consent for the methotrexate and pulses randomisation (R2) **as soon as possible after receiving the day 29 MRD result.** However, the patient should not be randomised until the receipt of the MRD result taken upon recovery from consolidation at week 14 (ALL patients).

Lymphoblastic Lymphoma

1. **At diagnosis**, provide parent/patient with the first (Registration) information sheet. Obtain consent for registration, trial entry and data collection. In addition this PIS will include details of any additional tests, procedures or samples for which informed consent is required but are not mandatory for entry into the UKALL 2011 trial e.g. sub studies.
2. **Then in good responders (patients with >35% reduction in tumour volume at the end of induction)**, provide parent/patient with the second (R2) information sheet and obtain consent for the methotrexate and pulses randomisation (R2) **as soon as possible after receiving the tumour volume assessment result at the end of induction.**

N.B. The methotrexate and pulses randomisation is a factorial design. For inclusion in the second part of the trial patients must consent to participate in **both** elements of the second randomisation (R2).

6. TRIAL ENTRY

Participating sites should be in receipt of a trial activation notification from CRCTU prior to any patients being registered/randomised to the trial.

Pre-treatment evaluations should be carried out by sites as detailed in section 5.1.

If informed consent is obtained, participants will be registered to the trial and assigned a trial specific number. It should be noted that there is no treatment randomisation at this stage. Participants who are subsequently eligible for R2 at the given time point should be randomised prior to the commencement of protocol treatment, providing informed consent has been obtained.

Due to closure of R1 (31-Mar-2017) all patients entering the trial at diagnosis will now be registered to the trial for data collection only (see section 9).

All patients should be registered to the trial within 7 days of commencement of induction treatment

6.1 Procedure for online patient registration and randomisation

- Registration for the trial should be performed by sites using the online electronic data capture system. Informed consent must be obtained prior to registration. Once an eligibility checklist has been completed, in order to register a patient for UKALL 2011, the online registration form must be completed.
- Additional informed consent for R2 must be obtained prior to the randomisation. Randomisation for R2 should also be performed by sites using the online remote data entry system at the protocol mandated time point. Informed consent for R2 must be obtained prior to the randomisation. In order to be able to randomise a patient for R2 you will be required to complete the paper R2 eligibility checklist to confirm that the patient is eligible for R2 before proceeding to randomise the patient using the electronic data capture system.

Registration and randomisation of patients can be achieved by logging on to:

<https://www.cancertrials.bham.ac.uk/UKALL2011Live>

The program will confirm eligibility and allocate treatment (R2) via a computerised minimisation algorithm, developed by the CRCTU.

A copy of each trial entry report should be printed and retained in the Investigator Site File (ISF) and patient's notes. At trial entry, this will include the patient's unique Trial Number (TNO). This number should be used on all serious adverse event (SAE) Forms and all correspondence relating to that patient.

A copy of the patient's Informed Consent Form and eligibility checklist must be sent in the post to the UKALL 2011 Trial office.

6.2 Emergency registrations and randomisations

In case of any problems with online randomisation, a paper eligibility checklist and randomisation form should be completed. These details can be phoned or faxed through to the UKALL 2011 Trials Office at the Cancer Research UK Clinical Trials Unit (CRCTU), University of Birmingham using the below numbers:

***REGISTRATION &
RANDOMISATION***

(09:00 to 17:00, Monday to Friday)

☎ 0121 414 7626 or ☎ 0121 415 1068

Fax: 0121 415 9520

7. TREATMENT DETAILS

7.1 Risks of intrathecal therapy

All medical staff involved in the care of patients with leukaemia and lymphoma MUST be aware that the **inadvertent** administration of vincristine by the **intrathecal** route is invariably **FATAL**. **Vincristine should NOT BE AVAILABLE when an intrathecal needle is *in situ*.** This protocol has been written to provide separation of intrathecal methotrexate administration from intravenous vincristine administration in time. An additional precaution is that the two drugs should not be administered in the same place.

All sites must ensure they comply with Department of Health HSC 2008/001 *"Updated national guidance on the safe administration of intrathecal chemotherapy"* or other country specific guidance and any subsequent amendments. The document can be downloaded from the Department of Health website here: [DH HSC 2008/001](#) (NB: HSC 2008/001 is due for review in August 2011.)

It is appreciated that not all centres will be able to administer the drugs in the suggested order within a given week, and therefore local factors may determine an alternative scheme. The single most crucial element in avoiding errors is the appropriate education and training of all personnel involved in the administration of chemotherapy.

7.2 General management of all treatment regimens

7.2.1 Registration

Treatment guidelines for induction and consolidation (registration) should be followed as closely as possible so that patients randomised to R2 have received uniform and standardised prior treatment..

7.2.2 Randomisation 2 (R2)

Randomised treatment (R2) as set out in this document must be followed as closely as possible. Where drug timings must be **strictly followed** this is specified in the treatment sections. The drug doses specified in this protocol must be given at full dosage at all times unless there is intolerance when the dosage must only be modified according to the instructions set out in this protocol. Guidelines for determining weight for dose calculation are given in section 7.4. In exceptional circumstances, the responsible clinician may decide in consultation with the Chief Investigator or Co-investigators that exact adherence is unjustifiably hazardous for the patient. Doses must be returned to that specified in this document thereafter and dosages must be regularly increased as the patient grows (see section 7.4 for further information on dosage calculations).

All drugs used in continuing therapy must be started at maximum dosage as specified in the protocol. In addition, in patients on daily mercaptopurine in whom no toxicity is evident, the mercaptopurine dose should be increased until toxicity occurs (see section 7.11.2).

Doses should be within 5% of the target dose. In the event of intolerance of the liquid preparation, alternate day tablet dosing can be employed to maintain 5% accuracy over 48 hours. No single dose should exceed 10% discrepancy from the target dose.

If the 12 week maintenance therapy cycle includes gaps when treatment has been stopped because of toxicity or infection the omitted oral mercaptopurine and methotrexate are counted as given and not made up; i.e. the clock does not stop when the drugs are stopped. In this way, cycles are maintained at 12 weeks; gaps in therapy and total dosage per cycle can be related to outcome. All significant dosage modifications and interruptions must be recorded on the CRF.

7.2.2.1 Administration of vincristine

Sites must ensure they comply with the requirements of the NHS National Patient Safety Agency Rapid Response Report NPSA/2008/RRR04 *"Using Vinca Alkaloid Minibags (Adult/Adolescent Units)"* or other country specific guidance for the administration of vincristine doses given throughout this

protocol. The document can be found here: http://www.nrls.npsa.nhs.uk/NPSA_2008_RRR04. This has been agreed by the MHRA to be part of the assembly process.

7.2.2.2 Extravasation

Vincristine, doxorubicin and daunorubicin are all vesicant drugs. Take care to avoid extravasation. Patients should be closely monitored for signs of extravasation during administration and follow local policy for treatment of extravasation. Incidents of extravasation should be reported and shared via the National Extravasation Information Service (www.extravasation.org.uk).

7.2.2.3 Missed oral doses

During R2 treatment oral drug doses must be taken as close as possible to the prescribed time. Missed or vomited doses should not be made up and the next dose should be taken at the scheduled time, unless the dose was vomited within 30 minutes of administration (according to local policy). Patients should be asked to report any missed doses at routine clinic visits.

7.2.2.4 Administration of intrathecal methotrexate

Intrathecal methotrexate should be given at a concentration of ≤ 2.5 mg/ml to ensure adequate distribution within the CSF. It is recommended that patients lie prone for 1 hour after administration.

7.2.2.5 Administration of intrathecal chemotherapy via Ommaya reservoir

If there are problems delivering intrathecal chemotherapy via lumbar puncture then intraventricular administration via an Ommaya reservoir may be an appropriate alternative. Intraventricular chemotherapy should be given according to the same schedule, but at 50% of the corresponding age-based doses that would be given by lumbar puncture. This dose reduction reflects the significantly higher CSF concentrations of intrathecal chemotherapy that occur after direct intraventricular instillation (47).

End of treatment

Registration

A minimal data set will be collected for registered patients (per section 9.0). Treatment data for registered patients that decline R2 treatment will continue until the completion of consolidation. In addition to this, registered patients should be followed up on an ad hoc basis for relapse, death and secondary malignancy only. Upon completion of treatment there will be a minimum 5 year follow up period.

R2

End of protocol treatment is 24 months for girls and 36 months for boys from the start of interim maintenance.

7.3 Trial Treatment

It should be noted that all treatment for registered patients will be designated NIMPs.

For all patients participating in R2, the treatment must be administered according the trial protocol. For these patients, the treatments designated as Investigational Medicinal Products (IMPs) and Non-Investigational Medicinal Products (NIMPs) are listed below.

Investigational Medicinal Products (IMPs)

The following drugs are Investigational Medicinal Products (IMPs) in this trial:

Phase of treatment	Regimen	IMP	Formulation
Standard Interim Maintenance	A,B	dexamethasone/prednisolone* vincristine mercaptopurine oral methotrexate intrathecal methotrexate***	Tablets, soluble tablets or syrup Injection Tablets or oral suspension Tablets or oral liquid Injection
Protocol M	A,B	mercaptopurine intravenous methotrexate intrathecal methotrexate***	Tablets or oral suspension Injection Injection
Capizzi Maintenance	C	vincristine intravenous methotrexate pegaspargase/crisantaspase** intrathecal methotrexate***	Injection Injection Injection Injection
Protocol M-A	C	mercaptopurine intravenous methotrexate intrathecal methotrexate*** pegaspargase/crisantaspase**	Tablets or oral suspension Injection Injection Injection
Maintenance	A,B,C	vincristine dexamethasone/prednisolone* intrathecal methotrexate***	Injection Tablets, soluble tablets or syrup Injection

NB: Only licensed products (including Oncaspar and 10mg mercaptopurine tablets which are currently only licensed in another European country) or approved IMP trial supplies may be used as IMPs. See Appendix 16 for details on IMP supplies.

** Where crisantaspase is used in place of pegaspargase (e.g. in patients allergic to pegaspargase), crisantaspase is also an IMP in that phase of treatment.

*** Where cytarabine or hydrocortisone is used in place of intrathecal methotrexate, cytarabine/hydrocortisone is also an IMP in that phase of treatment.

Use of intravenous dexamethasone is permissible in severely ill patients.

7.3.1 Non-Investigational Medicinal Products (NIMPs)

The following drugs are Non-Investigational Medicinal Products (NIMPs) in this trial:

Phase of treatment	Regimen	NIMP
Standard Induction	A,B,C	dexamethasone/prednisolone* vincristine daunorubicin pegaspargase/crisantaspase** intrathecal methotrexate*** mercaptopurine
Consolidation (all drugs are NIMPs)	A	intrathecal methotrexate*** mercaptopurine
BFM Consolidation (all drugs are NIMPs)	B	cyclophosphamide cytarabine mercaptopurine intrathecal methotrexate***
Augmented BFM consolidation (all drugs are NIMPs)	C	cyclophosphamide cytarabine mercaptopurine vincristine pegaspargase/crisantaspase** intrathecal methotrexate***
Delayed Intensification (all drugs are NIMPs)	A,B,C	dexamethasone/prednisolone* vincristine doxorubicin pegaspargase/crisantaspase** intrathecal methotrexate*** cyclophosphamide mercaptopurine cytarabine
Maintenance	A,B,C	mercaptopurine oral methotrexate

Licensed products should be used as NIMPs, however unlicensed 'Specials' may be used as NIMPs (when an unlicensed product is deemed the most appropriate for the patient) if purchased from a Specials manufacturer. It is recommended that if using the 'Special' formulation of oral methotrexate liquid that the formulation is purchased from the same manufacturer as the equivalent IMP formulation for this trial. See Appendix 16 and the Pharmacy Manual for further details.

In addition, all supportive care (e.g. fluids, allopurinol, co-trimoxazole) and rescue medications (e.g. folinic acid) are NIMPs.

* Where prednisolone is used in place of dexamethasone (e.g. due to severe toxicity), prednisolone is also a NIMP in that phase of treatment.

** Where crisantaspase is used in place of pegaspargase (e.g. in patients allergic to pegaspargase), crisantaspase is also a NIMP in that phase of treatment.

*** Where cytarabine or hydrocortisone is used in place of intrathecal methotrexate, cytarabine/hydrocortisone is also an NIMP in that phase of treatment.

7.4 Determining weight for dosage calculations

7.4.1 Children (patients aged under 18 years)

All children should be weighed in light weight clothing and scales should be calibrated regularly. To ensure that children are treated effectively, without overdosing due to treatment related fat deposition, the Body Mass Index (BMI) should be checked at diagnosis, prior to interim maintenance and at the beginning of each cycle in maintenance.

Calculate using the following formula:

$$\text{BMI} = \text{weight (kg)} / \text{height}^2 \text{ (mxm)}$$

The BMI can then be compared to the standard Child Growth foundation BMI charts for the appropriate sex.

Use the BSA charts in children⁶ to determine the surface area for dose calculation. These can be found at the back of the BNF for Children.

For children with a BMI that falls within the 2nd-98th percentiles, dose by **actual weight** using the BSA charts to determine the surface area (SA) for dose calculation. These weights should be taken:-

- 1) At diagnosis
- 2) Prior to interim maintenance
- 3) At the beginning of each maintenance cycle

For children who have a BMI >98th percentile read off the BMI at 98th percentile for their age. Child Growth Foundation charts can be downloaded from the Royal College of Paediatrics and Child Health at www.rcpch.ac.uk.

Calculate the **dosing weight** using the formula:

$$\text{Dosing weight (kg)} = \text{BMI at 98}^{\text{th}} \text{ percentile} \times \text{Ht}^2 \text{ (mxm)}$$

For children <2nd percentile, repeat as above reading the BMI at the 2nd percentile for calculation.

For children who have greater than 10% weight loss due to illness during treatment it may be necessary to re- assess their BMI and dosing weight prior to recommencing treatment.

Doses should be rounded to the nearest half tablet for children taking tablets.

7.4.1.1 Rationale for frequency of weighing for determination of changes in drug dosage.

A normal child aged 2-10 years gains only 2-6kg/year¹. This equates to an average of 1kg every 3 months (maximum 1.5kg).

If children are weighed fully clothed rather than in light weight clothing, the extra weight will be greater than the quarterly change in weight. Scales, even regularly calibrated, also carry a measurement error. These errors, coupled with weight variation due to physical, endocrine, nutritional, and psychosocial effects make repeated measurements inaccurate².

Children having repeat doses of steroids will have greater than average weight gain, but this is primarily deposited as fat- particularly truncal fat³. Unless drug treatment is with very lipophilic drugs the weight gain should not affect the therapeutic doses required to treat ALL. Only daunorubicin,

doxorubicin and vincristine have longer T1/2 and larger volume of distribution, indicating major transfer to body compartments other than the blood. Both anthracyclines are ionic compounds and they and their active metabolites are highly protein bound, which would suggest less deposit into fat than into lean mass. Vincristine is highly water soluble and readily distributed into tissues. As it is predominantly renally excreted it is again unlikely that it is highly deposited in fat⁴.

Do not adjust doses on the basis of weight change any more frequently than every 3 months. In practice the changes should equate to:

- 1) At diagnosis
- 2) Prior to interim maintenance
- 3) At the beginning of each maintenance cycle

The BSA charts for children have been validated for children with an understanding that they are less accurate at the extremes of obesity and underweight. There is no defined cut off. There is no defined BMI for obesity in children, but children with BMI >98th percentile for age have been shown to have an increased risk of related morbidity in adult life⁵. The Child growth foundation BMI charts for boys and girls mark the 2nd and 98th percentiles so it would logical to use these cut off points

Doses have traditionally been based on total weight rather than lean weight, so to change to lean body weight dosing could result in under-treatment. Dexamethasone is likely to cause greater weight gain than prednisolone¹. This may be compounded by the secular changes in UK society. It would therefore seem prudent to have a defined maximum BMI percentile, for dosing; weight percentiles taking no account of tall or small stature children.

REFERENCES

1. Clinical Paediatric endocrinology. Brook and Hindmarsh.
2. Short term growth: rhythms, chaos or noise? Wales J et al Archives Dis Child 1994 71: 84-89
3. Bodyweight change as an adverse effect of drug treatment- Mechanisms and management Pijl H et al Drug Safety 1996 14(5); 329-42
4. Micromedex Drug database September 2004
5. Use of weight for height indices in children to predict adult overweight: the Bogalusa Heart Study Int J Obesity 1996 21:715-21
6. Sharkey et al. Body surface area estimation in children using weight alone: application in paediatric oncology. Br J Cancer 2001; 85: 23–8.

7.4.2 Adults (patients aged 18 and above)

Body surface area for adults should be calculated using the DuBois formula:

$$\text{Body Surface Area (m)} = 0.007184 \times \text{weight (kg)}^{0.425} \times \text{height (cm)}^{0.725}$$

Weight should be taken at the following time points for calculation:

- 1) At diagnosis
- 2) Prior to interim maintenance
- 3) At the beginning of each maintenance cycle

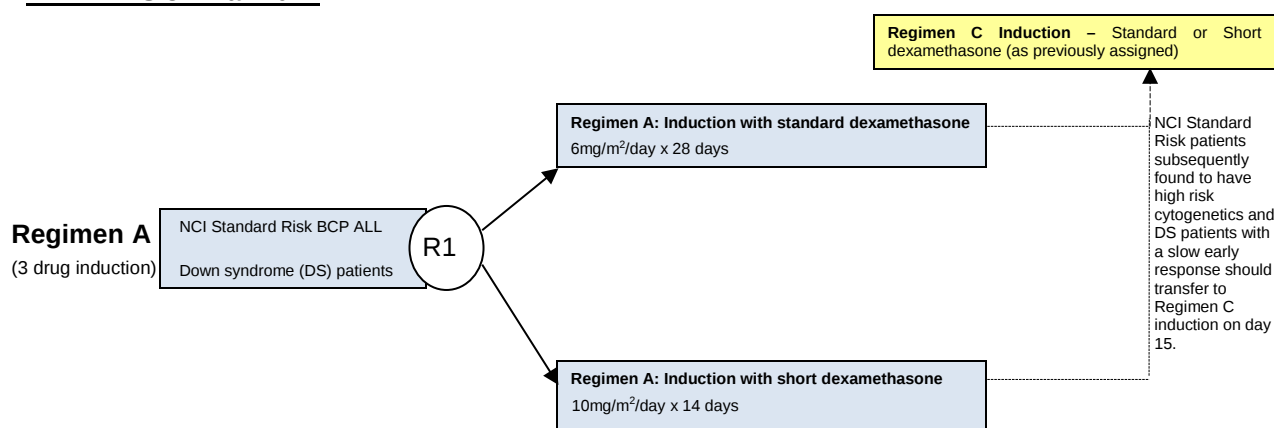
Body surface area should be recalculated with any major weight change as per local practice. It is recommended that sites follow local guidelines for dose adjustments for obese patients.

7.5 Regimen A Treatment Schedule

7.5.1 Summary of Regimen A Treatment

TREATMENT PHASE	DURATION	DETAILS OF TREATMENTS/RANDOMISATIONS
Induction	5 weeks (weeks 1-5)	It is recommended that patients registered on the study should be treated according to standard practice including dexamethasone 6mg/m ² /day orally for 28 days and standard MRD stratified post induction treatment.
Consolidation	3 weeks (weeks 6-8)	
Interim Maintenance	9 weeks (weeks 9-17)	Randomisation 2 (R2): standard interim maintenance vs. high dose methotrexate (Protocol M)
Delayed Intensification	7 weeks (weeks 18-24)	
Maintenance	Continues for exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance in ALL and LBL.	Randomisation 2 (R2): vincristine and dexamethasone pulses vs. no pulses.

Figure 6: Allocation of induction treatment in Regimen A – TREATMENT ARM CLOSED TO NEW PATIENTS 31-Mar-2017



7.5.2 Regimen A: Induction

It is recommended that patients registered on the study should be treated according to the standard arm with dexamethasone 6mg/m²/day orally for 28 days and standard MRD stratified post induction treatment.

This regimen is for patients with NCI Standard Risk BCP ALL and Down syndrome (DS) patients.

This regimen is not for patients with high risk cytogenetics. Patients known to have high risk cytogenetics at the start of treatment should receive regimen B induction.

Patients receiving Regimen A induction must be before their 10th birthday at diagnosis and have a highest white cell count before starting treatment of <50x10⁹/L.

This phase runs for 5 weeks from day 1 (beginning of week 1) to day 35 inclusive (end of week 5).

N.B Patients subsequently found to have high risk cytogenetics and DS patients with a slow early response should transfer to Regimen C Induction at day 15

Patients with CNS disease at diagnosis

Patients with CNS disease at diagnosis should receive weekly intrathecal methotrexate obtained 4 doses during induction. See section 3.7.1 for details.

Use of intravenous dexamethasone in severely ill patients

Severely ill patients requiring intravenous dexamethasone at the beginning of induction will not be excluded from randomisation. Intravenous dexamethasone should be given at the randomised dose and switched to oral therapy as soon as clinical circumstances allow.

a)	Fluids	All patients should be adequately hydrated (at least 2-2.5 L/m ² /24hrs). Given parenterally for the first 48 hours.
b)	Allopurinol	100mg/m ² oral three times daily Should be started 24 hours before chemotherapy and continue for 5 days. NB. Maximum recommended dose in children <15 years old is 400mg/day. Alternative therapy can be used according to local practice.
c)	Dexamethasone	All patients should receive dexamethasone starting on day 1. Oral dexamethasone 6mg/m ² /day (maximum dose 10mg/day in induction only) for 28 days starting on day 1 and then tapered over the next 7 days. The steroid should be divided into two doses per day.
	Dexamethasone contd.	For Down Syndrome patient aged ≥10 years: Oral dexamethasone 10mg/m ² /day on days 1-7 and 15-21 no taper. The steroid should be divided into two doses per day. Do not cap dexamethasone dose. NB. For severely ill patients, it is permissible to use intravenous dexamethasone.

d)	Vincristine	1.5mg/m ² (maximum single dose 2mg) intravenous weekly for five weeks starting on day 2 and continuing on days 9, 16, 23 and 30.																				
e)	Pegaspargase (Oncaspar)	1000iu/m ² intramuscular on day 4 and day 18																				
f)	Intrathecal methotrexate	On days 1, 8⁺ and 29. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. NB Patients who have CNS disease at presentation should receive weekly doses until two clear CSF samples are obtained (see section 3.7.1). Do not schedule vincristine on the same day as the intrathecal methotrexate + Regimen A patients can receive day 8 intrathecal methotrexate with a day 15 bone marrow, if appropriate																				
g)	Mercaptopurine	75mg/m ² /day orally once a day starting on day 29 (beginning week 5) (if neutrophils >0.75x10 ⁹ /L and platelets >75x10 ⁹ /L) and continuing to day 21 of consolidation (4 weeks from the start in week 5 of induction). If necessary give extra doses between induction and consolidation to ensure continuity of therapy. Dose adjustments are described in section 7.11.2.																				
h)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	This drug is given as PCP prophylaxis orally twice a day (bd) on 2 consecutive days each week starting from day 1. Dose for children is based on surface area as detailed below: <table><tr><th>Surface area</th><th>Co-trimoxazole</th><th>Trimethoprim</th><th>Sulphamethoxazole</th></tr><tr><td>0.5-0.75m²</td><td>240mg bd</td><td>40mg bd</td><td>200mg bd</td></tr><tr><td>0.76-1.0m²</td><td>360mg bd</td><td>60mg bd</td><td>300mg bd</td></tr><tr><td>1.1m² – 1.5m²</td><td>480mg bd</td><td>80mg bd</td><td>400mg bd</td></tr><tr><td>*Over 1.5 m²</td><td>480-960mg</td><td>80-120mg bd</td><td>400-800mg bd</td></tr></table> *according to local policy over 1.5m² See also section 7.11.3 for details of alternative PCP prophylaxis regimens and permitted dose modifications for toxicity.	Surface area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole	0.5-0.75m ²	240mg bd	40mg bd	200mg bd	0.76-1.0m ²	360mg bd	60mg bd	300mg bd	1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd	*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd
Surface area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole																			
0.5-0.75m ²	240mg bd	40mg bd	200mg bd																			
0.76-1.0m ²	360mg bd	60mg bd	300mg bd																			
1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd																			
*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd																			

TESTS	Bone Marrow for MRD - <u>ALL PATIENTS ONLY</u>	Bone marrow should be taken at diagnosis and on days 8, 15* and 29. - Send day 29 bone marrow for MRD - Day 8/15 bone marrow is used to assess RER/SER. * If RER at day 8, sample at day 15 is not required.
	Study Closed to recruitment on 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Bone Marrow in ACD tube at diagnosis Peripheral blood in EDTA tube on days 9,16 & 30 See Appendix 12 for details of the samples required.
	Study Closed to recruitment on 11-Jan-2018 Dexamethasone Study samples (optional – consent must have been provided) - <u>ALL PATIENTS ONLY</u>	<u>All patients</u> Pre-treatment: saliva and blood sample Day 1: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 1. <u>Patients randomised to short dexamethasone (with continuous dosing on days 1-14)</u> Day 14: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 14. <u>Patients randomised to short dexamethasone (with split dosing on days 1-7 and 15-21)</u> Day 21: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 21. <u>Patients randomised to standard dexamethasone</u> Day 28: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 28. Appendix 15 – Dexamethasone pharmacokinetic study - CLOSED for details of the samples required
	<u>Study closed to recruitment on 31-Mar-2018</u> Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Samples</u> Blood samples taken before and after the first vincristine dose on day 2. Saliva sample taken for genetic analysis prior to vincristine PK sampling. <u>Physiotherapy Assessment</u> To be performed at diagnosis. See Appendix 24 for details of the samples required.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 1 should already have been completed (after informed consent has been obtained ideally prior to randomisation). Must be completed by end of Week 1 at the latest.

		QuEST and Quality of Life Questionnaire 2 must be completed at the end of induction and within 2 weeks of this time point at the latest.
--	--	---

7.5.3 Regimen A: Post-induction therapy

This regimen is for patients with **NCI Standard Risk BCP ALL**, without high risk cytogenetics, who are **MRD Low Risk** at day 29. This regimen should also be used in **NCI Standard Risk BCP ALL** patients who are **MRD NO result** and show a rapid early response “**rapid early responders**” at day 15 of induction without high risk cytogenetics.

NCI Standard Risk BCP ALL with persistent CNS disease or M2 marrow at the end of induction should transfer to Regimen C.

All patients apart from Down syndrome patients are eligible for the factorial methotrexate and pulses randomisation.

Figure 7: Allocation of post-induction treatment in Regimen A

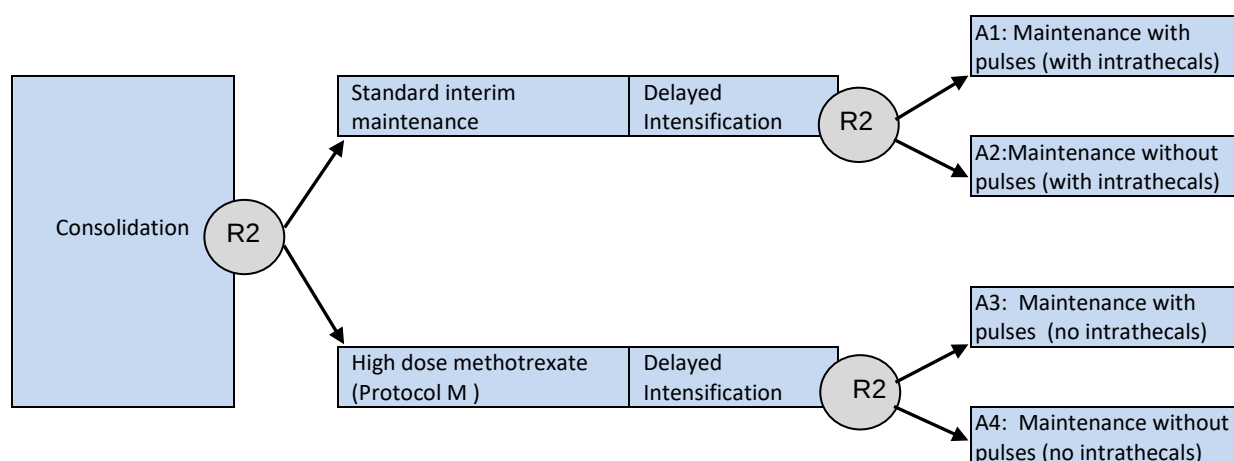


Figure 7

Allocation of Regimen A post-induction therapy. Patients receiving Regimen A are randomised (R2) into four groups to receive standard interim maintenance or high dose methotrexate (Protocol M) followed by a single delayed intensification and either maintenance therapy with pulses or maintenance therapy without pulses.

7.5.4 Regimen A: Consolidation

This phase runs for 3 weeks from day 1 (beginning of week 6) to day 21 inclusive (end of week 8).

Patients should have an absolute neutrophil count (ANC) $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase.

There are no IMPs in this treatment phase.

a)	Intrathecal methotrexate	On days 1, 8 and 15. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg.
b)	Mercaptopurine	75mg/m ² /day orally once a day starting on day 1 and continuing to day 21 (end of week 8) Dose adjustments are described in section 7.11.2.
c)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

	<u>Study closed to recruitment on 31-Mar-2018</u> Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed during weeks 6-8, following administration of the first 5 vincristine doses.
--	--	--

7.5.5 Regimen A: Standard Interim Maintenance

This regimen is for patients randomised to standard interim maintenance only.

This regimen is standard treatment for Regimen A patients and should be considered for those patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

This phase runs for 9 weeks from day 1 (beginning of week 9) to day 63 inclusive (end of week 17).

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase.

The IMPs in this treatment phase are highlighted in the table below.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally in 2 divided doses each day on days 1-5 (week 9) and days 29-33 (week 13).
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on day 1 (week 9) and day 29 (week 13).
c)	Mercaptopurine (THIS IS AN IMP)	75mg/m ² /day orally once a day on days 1-56 (weeks 9-16) but not days 57-63 (week 17) . Dose adjustments are described in section 7.11.2
d)	Oral methotrexate (THIS IS AN IMP)	20mg/m ² orally once per week during weeks beginning on days 1 (week 9), 8 (week 10), 22 (week 12), 29 (week 13), 36 (week 14), 50 (week 16) and 57 (week 17). Note none is given day 15 (week 11) and day 43 (week 15) as an intrathecal dose is given during that week. Oral methotrexate should be given as a single dose taken with mercaptopurine. Dose adjustments are described in section 7.11.2
e)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 (week 11) and 43 (week 15). Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine on the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.
-----	-------------------------------	---

7.5.6 Regimen A: High Dose Methotrexate (Protocol M)

This regimen is for patients randomised to high dose methotrexate (Protocol M) only

The administration of high dose methotrexate (HDMTX) carries significant potential toxicity which in extreme cases can be fatal. This is prevented by effective folinic acid rescue. Absolute compliance with the schedule of hydration, monitoring of levels and folinic acid rescue is mandatory in this phase of the protocol. It is vital to read this section of the protocol and Appendix 7 fully before commencing administration of HDMTX. Please also ensure that you have made certain that your patient has no contraindication to HDMTX as detailed in section 0. Please note that to be eligible for entry into this phase renal function should be within normal limits corrected for age.

This phase runs for 9 weeks from day 1 (beginning of week 9) to day 63 inclusive (end of week 17).

Prior to starting the first pulse of HDMTX patients must be free of infection, diarrhoea and mucositis and should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$. Renal function should be within normal limits corrected for age.

Prior to starting each subsequent pulse of HDMTX serum creatinine must be $<1.5 \times$ baseline or GFR creatinine clearance $>65\text{mL/minute}/1.73\text{m}^2$. If renal function does not recover, omit MTX. Do not give HDMTX to a patient with this degree of renal impairment, assuming that prolonged excretion can be managed with Carboxypeptidase. Counts permitting continue with mercaptopurine regardless of renal function throughout protocol M/M-A to 2 weeks after last course of HD MTX. ALT and AST should be less than 5 x normal. Wherever possible avoid concurrent NSAIDs, aminoglycosides and other nephrotoxic drugs. See Appendix 6 for information on drug interactions.

Prior to starting each subsequent pulse of HDMTX ANC should be $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$. If blood counts are not adequate on the scheduled day of infusion then stop 6-MP. On recovery to ANC $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$ then administer the scheduled dose of methotrexate and restart 6-MP at full $25\text{mg}/\text{m}^2$ dose.

The IMPs in this treatment phase are highlighted in the table below.

a)	Mercaptopurine (THIS IS AN IMP)	25mg/m ² /day orally once a day on days 1-56 (weeks 9-16) but not days 57-63 (week 17) . Do NOT adjust dose.
b)	Methotrexate (THIS IS AN IMP)	5 g/m ² intravenous on day 8, 22, 36 and 50. All patients must be prehydrated as per guideline in Appendix 7. The initial 10% of dose is delivered over 30 minutes, the remaining 90% over 23 ½ hours. NB method and schedule of administration should be guided by Appendix 7. Timings of drug administration must be followed strictly.
c)	Folinic acid	15mg/m ² intravenously at 42, 48 and 54 hours after the start of each methotrexate infusion. Do not cap the dose. Further details of the folinic acid rescue are given in Appendix 7. Timings of drug administration must be followed strictly.
d)	Intrathecal methotrexate (THIS IS AN IMP)	Should optimally be given within 2 hours after the start of methotrexate infusion on days 8, 22, 36 and 50. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs:12mg.

e)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Must be omitted during Protocol Re-start after day 53 when folinic acid rescue is complete.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.

7.5.7 Regimen A: Delayed Intensification

This phase runs for 7 weeks from day 1 (beginning of week 18) to day 49 inclusive (end week 24).

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. Once begun, therapy during weeks 18 – 21 is not interrupted for myelosuppression alone. Therapy due on day 1, week 22 should be delayed until ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ and once begun should not be interrupted solely for myelosuppression.

Treatment may be interrupted for **serious** infection (presumed or proven) such as Varicella, pneumocystis pneumonia, or neutropenia with fever.

There are no IMPs in this treatment phase.

Part 1, weeks 18-21		
a)	Dexamethasone	10mg/m ² /day orally for 7 days on days 2-8 (week 18) and 16-22 (week 20). No taper. Divide into two daily doses.
b)	Vincristine	1.5mg/m ² (maximum single dose 2mg) intravenous on days 2 (week 18), 9 (week 19) and 16 (week 20).
c)	Doxorubicin	25mg/m ² intravenous on days 2 (week 18), 9 (week 19) and 16 (week 20) given over a period of 1 hour (or as a bolus alongside a fast running infusion in adults)..
d)	Pegasparase (Oncaspar)	1000 iu/m ² intramuscular on day 4 (week 18)
e)	Intrathecal methotrexate	On day 1 (week 18). Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine on the same day as intrathecal methotrexate.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be given throughout this phase of treatment as per guidelines provided in induction regimen section.
Part 2, weeks 22-24		
Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase.		
g)	Cyclophosphamide	1,000mg/m ² intravenous over 20-30 min on day 29 (week 22). Give 125 ml/m ² /hr of Dextrose/Saline infusion for 30 minutes before the cyclophosphamide and for 3.5 hours afterwards, i.e. 4 hours in total. Mesna is not needed.
h)	Mercaptopurine.	All patients receive 6-MP 60mg/m²/day during delayed intensification. The drug is given once a day by mouth for 14

		days from day 29 (beginning of week 22) to day 42 (end of week 23). No dose adjustments, other than for toxicity are made during this block.
i)	Cytarabine	75 mg/m ² /day by intravenous or subcutaneously. Give on 4 consecutive days in weeks 22 and 23 starting on the same day each week. Cytarabine pulses should ideally start the day after cyclophosphamide when both drugs are given in the same week.
j)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Study closed to recruitment on 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood in EDTA tube Day 16 (12-18). See Appendix 12 for details of the sample required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed during weeks 22-24 of delayed intensification and before the onset of maintenance.

7.5.8 Regimen A: Maintenance

Note regarding randomisations. As per Figure 8 four maintenance regimens will be employed in UKALL 2011. All four employ daily oral mercaptopurine and weekly oral methotrexate. Patients randomised to receive high dose methotrexate in interim maintenance (Protocol M) do not receive continuing intrathecal therapy in maintenance. Patients randomised to receive “no pulses” do not receive monthly pulses of vincristine and dexamethasone.

Maintenance runs from day 1 cycle 1 (beginning of week 25). The duration of each cycle is 12 weeks. Treatment is stopped exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance/Protocol M for both ALL and LBL. The cycle in progress is stopped when this date is reached. No allowances are made for interruptions in treatment after the start of interim maintenance.

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. Once maintenance is started the clock does not stop regardless of whether treatment is given or not. Only mercaptopurine and oral methotrexate will be interrupted for myelosuppression and the time is not be made up. Vincristine and dexamethasone pulses (if given) should be given regardless of blood count. Days off therapy for intercurrent infections are counted as days of maintenance and the time is not made up.

Anaemia occurring in the course of maintenance therapy should be treated with transfusion and the dose of drug maintained. If *persistent* (> 4 weeks) *anaemia* occurs (i.e., haemoglobin below 8 g/dl) investigate for Parvovirus infection. Please contact the Chief Investigator or Co-investigators for advice.

Figure 8: Maintenance treatments in Regimen A

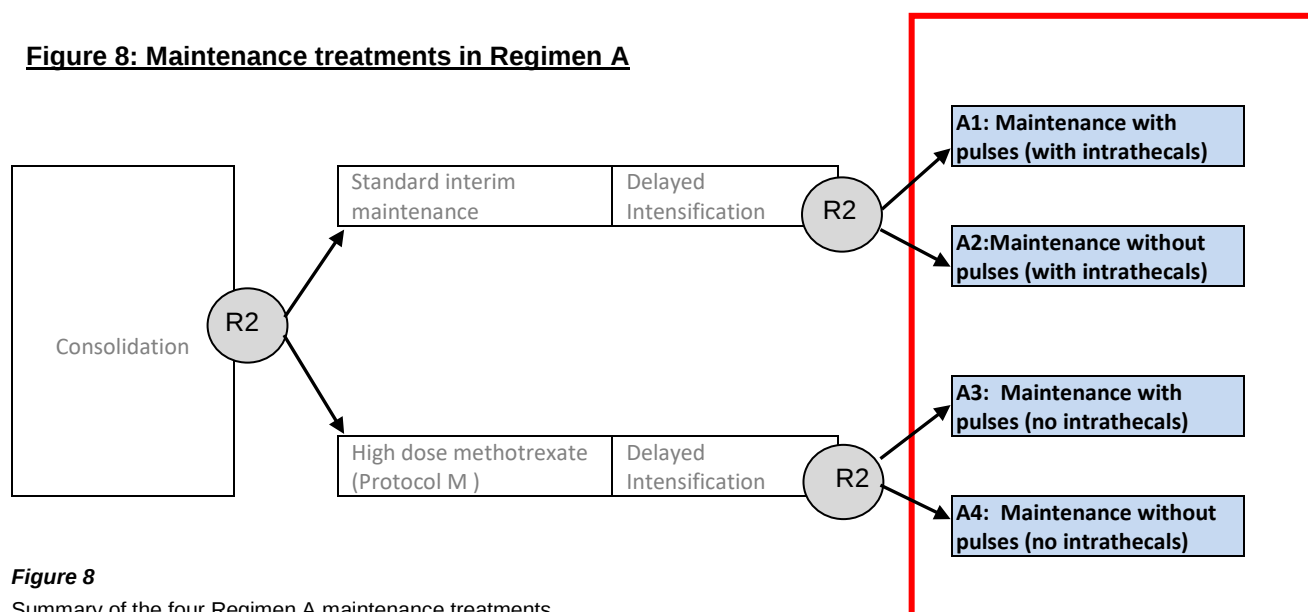


Figure 8

Summary of the four Regimen A maintenance treatments.

7.5.8.1 Maintenance Regimen A1

This regimen is for patients randomised to receive standard interim maintenance and vincristine/dexamethasone pulses only.

This regimen is standard treatment for Regimen A patients and should be considered for those patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

The IMPs in this treatment phase are highlighted in the table below.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses on days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2
d)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine for the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.
TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL</u> <u>PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 29, 53 and 77 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.5.8.2 Maintenance Regimen A2

This regimen is for patients randomised to receive standard interim maintenance and NO vincristine/dexamethasone pulses only.

The IMPs in this treatment phase are highlighted in the table below.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
c)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
d)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L.
	Asparaginase Study samples (optional – consent must have been provided)	See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018	<u>Physiotherapy Assessment</u> To be performed at weeks 29, 53 and 77 of maintenance.
	Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.5.8.3 Maintenance Regimen A3

This regimen is for patients randomised to receive high dose methotrexate (Protocol M) and vincristine/dexamethasone pulses only.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses on days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate	20mg/m ² orally once per WEEK throughout maintenance. Dose adjustments are described in section 7.11.2.
e)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L.
	Asparaginase Study samples (optional – consent must have been provided)	See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018	<u>Physiotherapy Assessment</u> To be performed at weeks 29, 53 and 77 of maintenance.
	Vincristine Study (optional – consent must have been provided) – <u>ALL</u> <u>PATIENTS ONLY</u>	To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.5.8.4 Maintenance Regimen A4

This regimen is for patients randomised to receive high dose methotrexate (Protocol M) and NO vincristine/dexamethasone pulses only.

There are no IMPs in this treatment phase.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per WEEK throughout maintenance. Dose adjustments are described in section 7.11.2.
c)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

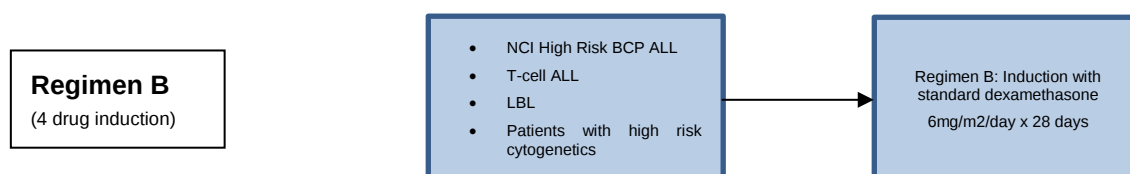
TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 29, 53 and 77 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.6 Regimen B Treatment Schedule

7.6.1 Summary of Regimen B Treatment

TREATMENT PHASE	DURATION	DETAILS OF TREATMENTS/RANDOMISATIONS
Induction	5 weeks (weeks 1-5)	It is recommended that patients registered on the study should be treated according to standard practice including dexamethasone 6mg/m ² /day orally for 28 days and standard MRD stratified post induction treatment.
Standard BFM Consolidation	5 weeks (weeks 6-10)	
Interim Maintenance	9 weeks (weeks 11-19)	Randomisation 2 (R2): standard interim maintenance vs. high dose methotrexate (Protocol M)
Delayed Intensification	7 weeks (weeks 20-26)	
Maintenance	Continues for exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance in ALL and LBL.	Randomisation 2 (R2): vincristine and dexamethasone pulses vs. no pulses.

Figure 9: Allocation of induction treatment in Regimen B



7.6.2 Regimen B: Induction

It is recommended that patients registered on the study should be treated according to the standard arm with dexamethasone 6mg/m²/day orally for 28 days and standard MRD stratified post induction treatment.

This regimen is for patients with NCI High Risk BCP ALL, all cases of T-cell ALL and LBL, and any patients known to have high risk cytogenetics at the start of their treatment (see section 3.7.4 for details).

All patients receive dexamethasone but the schedule differs according to randomisation and age.

Patients with BCP ALL receiving Regimen B induction must be aged ≥ 10 years at diagnosis and/or have a highest white cell count before starting treatment of $\geq 50 \times 10^9/L$.

This phase runs for 5 weeks from day 1 (beginning of week 1) to day 35 inclusive (end of week 5).

Patients with CNS disease at diagnosis

Patients with CNS disease at diagnosis should receive weekly intrathecal methotrexate until two consecutive clear CSFs have been obtained. See section 3.7.1 for details.

a)	Fluids	All patients should be adequately hydrated (at least 2-2.5 L/m ² /24hrs). Given parenterally for the first 48 hours.
b)	Allopurinol	100mg/m ² oral three times daily. Should be started 24 hours before chemotherapy and continue for 5 days. NB. Maximum recommended dose in children <15 years old is 400mg/day. Patients with very high white blood cell count ($>100 \times 10^9/L$), LBL or bulky T-cell disease are at risk of tumour lysis syndrome and rasburicase should be considered as an alternative in these patients.
c)	Dexamethasone	All patients should receive dexamethasone starting on day 1. Oral dexamethasone 6mg/m ² /day orally (maximum dose 10mg/day in induction only) for 28 days starting on day 1 and then tapered over the next 7 days. The steroid should be divided into two doses per day.
d)	Vincristine	1.5mg/m ² (maximum single dose 2mg) intravenous weekly for five weeks starting on day 2 and continuing on days 9, 16, 23 and 30.
e)	Daunorubicin	25mg/m ² intravenous over 1 hour (or as a bolus alongside a fast running infusion in adults) on days 2, 9, 16 and 23. NB: Daunorubicin is included in induction in Regimen B and this dosage is different to that recommended in Regimen C.
f)	Pegaspargase (Oncaspar)	1000 iu/m ² intramuscular on day 4 and day 18.

g)	Intrathecal methotrexate	On days 1, 8 and 29. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. NB Patients who have CNS disease at presentation should receive weekly doses until two clear CSF samples are obtained (see section 3.7.1) Do not schedule vincristine on the same day as the intrathecal methotrexate.																				
h)	Mercaptopurine	60mg/m²/day orally once a day starting on day 29 (beginning week 5) (if neutrophils >0.75 x10⁹/L and platelets >75 x10⁹/L) and continuing to day 28 of consolidation. (5 weeks from the start in week 5 of induction). If necessary give extra doses between induction and consolidation to ensure continuity of therapy. . Do not adjust dose according to blood count. NB: the mercaptopurine dose in Regimen B induction is different to that given in Regimen A.																				
i)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	This drug is given as PCP prophylaxis orally twice a day (bd) on 2 consecutive days each week starting from day 1. Dose for children is based on surface area as detailed below: <table><tr><th>Surface area</th><th>Co-trimoxazole</th><th>Trimethoprim</th><th>Sulphamethoxazole</th></tr><tr><td>0.5-0.75m²</td><td>240mg bd</td><td>40mg bd</td><td>200mg bd</td></tr><tr><td>0.76-1.0m²</td><td>360mg bd</td><td>60mg bd</td><td>300mg bd</td></tr><tr><td>1.1m² – 1.5m²</td><td>480mg bd</td><td>80mg bd</td><td>400mg bd</td></tr><tr><td>*Over 1.5 m²</td><td>480-960mg</td><td>80-120mg bd</td><td>400-800mg bd</td></tr></table> *according to local policy over 1.5m² See also section 7.11.3 for details of alternative PCP prophylaxis regimens and permitted dose modifications for toxicity.	Surface area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole	0.5-0.75m ²	240mg bd	40mg bd	200mg bd	0.76-1.0m ²	360mg bd	60mg bd	300mg bd	1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd	*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd
Surface area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole																			
0.5-0.75m ²	240mg bd	40mg bd	200mg bd																			
0.76-1.0m ²	360mg bd	60mg bd	300mg bd																			
1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd																			
*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd																			

TESTS	Bone Marrow for MRD (mandatory) - <u>ALL PATIENTS ONLY</u>	Bone marrow should be taken at diagnosis and on days 8 and 29. - Send day 29 bone marrow for MRD. - Day 8 bone marrow is used to assess RER/SER.
	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Bone Marrow in ACD tube at diagnosis Peripheral blood in EDTA tube on day 9,16 & 30 See Appendix 12 for details of the samples required.
	Closed to recruitment 11-Jan-2018 Dexamethasone Study samples (optional – consent must have been provided)	<u>All patients</u> Pre-treatment: saliva and blood sample Day 1: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 1. <u>Patients randomised to short dexamethasone (with continuous dosing on days 1-14)</u>

	been provided) - <u>ALL PATIENTS ONLY</u>	Day 14: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 14. <u>Patients randomised to short dexamethasone (with split dosing on days 1-7 and 15-21)</u> Day 21: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 21. <u>Patients randomised to standard dexamethasone</u> Day 28: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 28. See Appendix 15 – Dexamethasone pharmacokinetic study - CLOSED for details of the samples required
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Samples</u> Blood samples taken before and after the first vincristine dose on day 2. Saliva sample taken for genetic analysis prior to vincristine PK sampling. <u>Physiotherapy Assessment</u> To be performed at diagnosis. See Appendix 24 for details of the samples required.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 1 should already have been completed (after informed consent has been obtained ideally prior to randomisation). Must be completed by end of Week 1 at the latest QuEST and Quality of Life Questionnaire 2 must be completed at the end of induction and within 2 weeks of this time point at the latest.

7.6.3 Regimen B: Post-induction therapy

This regimen is for the following patients:

- 1) Patients with **NCI High Risk** BCP ALL and all T-ALL without high risk cytogenetics as follows:
 - 1) those who are **MRD Low Risk** at day 29
 - 2) those with **MRD NO result** who show a rapid early response “**rapid early responders**” at day 8 of induction.
- 2) LBL patients with B-cell precursor immunophenotype without high risk cytogenetics

Patients with persisting CNS disease, high risk cytogenetics or M2 marrow at the end of induction should transfer to Regimen C.

All patients are eligible for the factorial methotrexate and pulses randomisation.

Figure10: Allocation of post-induction treatment in Regimen B

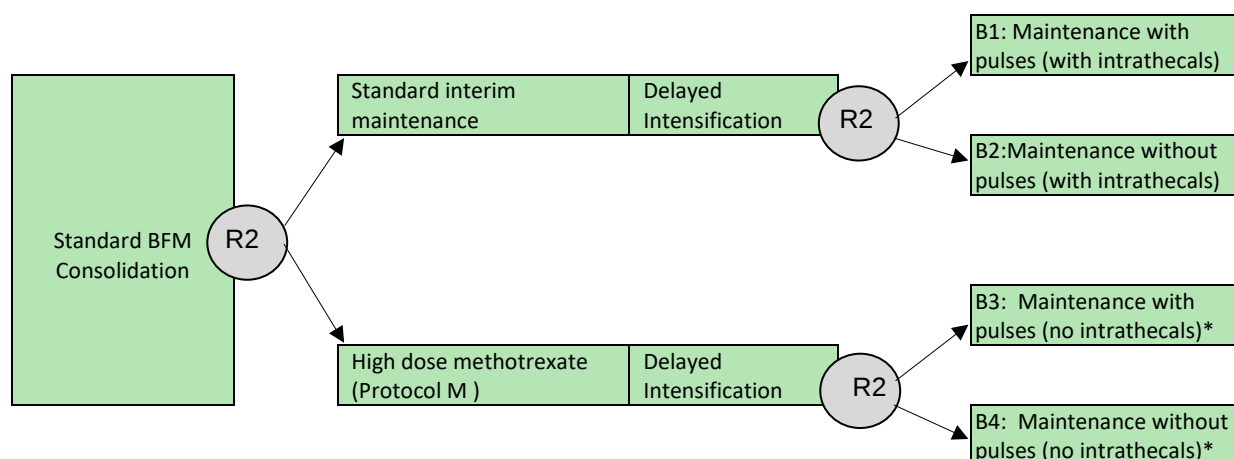


Figure 10

Allocation of Regimen B post-induction therapy. Patients receiving Regimen B are randomised (R2) into four groups to receive standard interim maintenance or high dose methotrexate (Protocol M) followed by a single delayed intensification and either maintenance therapy with pulses or maintenance therapy without pulses

* T-ALL patients with a presenting WCC >100x10⁹/L receive continuing intrathecal therapy.

7.6.4 Regimen B: Standard BFM Consolidation

This phase runs for 5 weeks from day 1 (beginning of week 6) to day 35 inclusive (end of week 10).

Patients should have an absolute neutrophil count (ANC) $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. If the blood count has not recovered and marrow is hypocellular M1, delay the start of consolidation one week and repeat the bone marrow to confirm M1 status. If blood counts had recovered to ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ at week 5 but subsequently fallen again, continue with week 6 therapy as long as patient clinically well.

Therapy during this phase should not be interrupted for myelosuppression.

There are no IMPs in this treatment phase.

a)	Cyclophosphamide	1000mg/m ² intravenous over 20-30 minutes on days 1 and 15. Give 125mls/m ² /hr of Dextrose/ Saline infusion for 30 minutes before starting cyclophosphamide and for 3.5 hours afterwards, i.e. 4 hours in total. Mesna is not needed.
b)	Cytarabine	75mg/m ² /day intravenous or subcutaneously. Give on 4 consecutive days in weeks 6, 7, 8 and 9 starting on the same day each week. Cytarabine pulses should ideally start the day after cyclophosphamide when both drugs are given in the same week.
c)	Mercaptopurine	Continue oral mercaptopurine 60mg/m ² /day once per day to day 28 of standard BFM consolidation (5 weeks in total from the start in week 5 of induction). Do not adjust dose according to blood count.
d)	Intrathecal methotrexate	On days 1, 8 and 15. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg.
e)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

Tests	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	Physiotherapy Assessment To be performed during weeks 6-8, following administration of the first 5 vincristine doses.
--------------	--	---

7.6.5 Regimen B: Standard Interim Maintenance

This regimen is for patients randomised to standard interim maintenance only.

This regimen is standard treatment for Regimen B patients and should be considered for those patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

This phase runs for 9 weeks from day 1 (beginning of week 11) to day 63 inclusive (end of week 19).

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase.

The IMPs in this treatment phase are highlighted in the table below.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses each day on days 1-5 (week 11) and days 29-33 (week 15)
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on day 1 (week 11) and day 29 (week 15).
c)	Mercaptopurine (THIS IS AN IMP)	75mg/m ² /day orally once per day on days 1-56 (weeks 11-18) but not days 57-63 (week 19) . Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate (THIS IS AN IMP)	20mg/m ² orally once per week during weeks beginning on days 1 (week 11), 8 (week 12), 22 (week 14), 29 (week 15) 36 (week 16), 50 (week 18) and 57 (week 19). Note none is given day 15 (week 13) or day 43 (week 17) as an intrathecal dose is given during these weeks. Oral methotrexate should be a single dose taken with mercaptopurine. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 (week 13) and day 43 (week 17). Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg Do not schedule vincristine on the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.

7.6.6 Regimen B: High Dose Methotrexate (Protocol M)

This regimen is for patients randomised to high dose methotrexate (Protocol M) only.

The administration of high dose methotrexate (HDMTX) carries significant potential toxicity which in extreme cases can be fatal. This is prevented by effective folinic acid rescue. Absolute compliance with the schedule of hydration, monitoring of levels and folinic acid rescue is mandatory in this phase of the protocol. It is vital to read this section of the protocol and Appendix 7 fully before commencing administration of HDMTX. Please also ensure that you have made certain that your patient has no contraindication to HDMTX as detailed in section 0. Please note that to be eligible for entry into this phase renal function should be within normal limits corrected for age.

This phase runs for 9 weeks from day 1 (beginning of week 11) to day 63 inclusive (end of week 19).

Prior to starting the first pulse of HDMTX patients must be free of infection, diarrhoea and mucositis and should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. Renal function should be within normal limits corrected for age.

Prior to starting each subsequent pulse of HDMTX serum creatinine must be $<1.5 \times$ baseline or GFR creatinine clearance $>65\text{mL/minute}/1.73\text{m}^2$. If renal function does not recover, omit MTX. Do not give HDMTX to a patient with this degree of renal impairment, assuming that prolonged excretion can be managed with Carboxypeptidase. Counts permitting, continue with mercaptopurine regardless of renal function throughout protocol M/M-A to 2 weeks after last course of HD MTX. ALT and AST should be less than 5 x normal. Wherever possible avoid concurrent NSAIDs, aminoglycosides and other nephrotoxic drugs. See Appendix 6 for information on drug interactions.

Prior to starting each subsequent pulse of HDMTX ANC should be $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$. If blood counts are not adequate on the scheduled day of infusion then stop 6-MP. On recovery to ANC $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$ then administer the scheduled dose of methotrexate and restart 6-MP at full 25mg/m^2 dose.

The IMPs in this treatment phase are highlighted in the table below.

a)	Mercaptopurine (THIS IS AN IMP)	25mg/m ² /day orally once a day on days 1-56 (weeks 11-18) but not days 57-63 (week 19). Do NOT adjust dose.
b)	Methotrexate (THIS IS AN IMP)	5g/m ² intravenous on day 8, 22, 36 and 50. All patients must be prehydrated as per guideline in Appendix 7. The initial 10% of dose is delivered over 30 minutes, the remaining 90% over 23 ½ hours. NB method and schedule of administration should be guided by Appendix 7. Timings of drug administration must be followed strictly.
c)	Folinic acid	15mg/m ² intravenous 42, 48 and 54 hours after the start of each methotrexate infusion. Further details of the folinic acid rescue are given in Appendix 7. Timings of drug administration must be followed strictly.
d)	Intrathecal	Should optimally be given within 2 hours after the start of

	methotrexate (THIS IS AN IMP)	methotrexate infusion on days 8, 22, 36 and 50. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg
e)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Must be omitted during Protocol M Re-start after day 53 when folinic acid rescue is complete.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.

7.6.7 Regimen B: Delayed Intensification

This phase runs for 7 weeks from day 1 (beginning of week 20) to day 49 inclusive (end week 26)

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. Once begun, therapy during weeks 20 – 23 is not interrupted for myelosuppression alone. Therapy due day 1, week 24 should be delayed until ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ and once begun should not be interrupted solely for myelosuppression.

Treatment may be interrupted for **serious** infection (presumed or proven) such as Varicella, pneumocystis pneumonia, or neutropenia with fever.

There are no IMPs in this treatment phase.

Part 1, weeks 20-23		
a)	Dexamethasone	10mg/m ² /day orally for 7 days on days 2- 8 (week 20) and 16-22 (week 22). No taper. Divide into two daily doses. Do not cap dexamethasone dose.
b)	Vincristine	1.5mg/m ² (maximum single dose 2mg) intravenous on days 2 (week 20), 9 (week 21) and 16 (week 22).
c)	Doxorubicin	25mg/m ² intravenous on days 2 (week 20), 9 (week 21) and 16 (week 22) given over a period of 1 hour (or as a bolus alongside a fast running infusion in adults).
d)	Pegaspargase (Oncaspar)	1000 iu/m ² intramuscular on day 4 (week 20)
e)	Intrathecal methotrexate	On day 1 (week 20). Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine on the same day as intrathecal methotrexate.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be given throughout this phase of treatment as per the guidelines provided in induction regimen section.
Part 2, weeks 24-26		
Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase		
g)	Cyclophosphamide	1,000mg/m ² intravenous over 20-30 min on day 29 (week 24). Give 125 mls/m ² /hr of dextrose/saline infusion for 30 minutes before the cyclophosphamide and for 3.5 hours afterwards, i.e. 4 hours in total. Mesna is not needed.
h)	Mercaptopurine.	All patients receive 6-MP 60mg/m²/day during delayed intensification. The drug is given once a day by mouth for 14 days from day 29 (beginning of week 24) to day 42 (end of week 25) inclusive. No dose adjustments other than for toxicity are made during this block.

i)	Cytarabine	75mg/m ² /day by intravenous or subcutaneously. Give on 4 consecutive days in weeks 24 and 25 starting on the same day each week. Cytarabine pulses should ideally start the day after cyclophosphamide when both drugs are given in the same week.
j)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral Blood in EDTA tube Day 16 (12-18). See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed during weeks 24-26 of delayed intensification and before the onset of maintenance.

7.6.8 Regimen B: Maintenance

Note regarding randomisations. As per Figure 11 four maintenance regimens will be employed in UKALL 2011. All four employ daily oral mercaptopurine and weekly oral methotrexate. **Patients randomised to receive high dose methotrexate in interim maintenance do not receive continuing intrathecal therapy except in patients with T-ALL and a presenting WCC $>100 \times 10^9/L$.** Patients randomised to receive “No pulses” do not receive monthly pulses of vincristine and dexamethasone.

Maintenance runs from day 1 cycle 1 (beginning of week 27). The duration of each cycle is 12 weeks. For all patients (ALL and LBL) treatment is stopped exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance/ Protocol M for both ALL and LBL. The cycle in progress is stopped when this date is reached. No allowances are made for interruptions in treatment after the start of interim maintenance.

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$ to start this phase. Once maintenance is started the clock does not stop regardless of whether treatment is given or not. Only mercaptopurine and oral methotrexate will be interrupted for myelosuppression and the time is not be made up. Vincristine and dexamethasone pulses (if given) should be given regardless of blood count. Days off therapy for intercurrent infections are counted as days of maintenance and the time is not made up.

Anaemia occurring in the course of maintenance therapy should be treated with transfusion and the dose of drug is maintained. If *persistent anaemia* occurs (i.e., haemoglobin below 8 g/dl) investigate for Parvovirus infection. Please contact the Chief Investigator or Co-investigators for advice.

Figure 11: Maintenance treatments in Regimen B

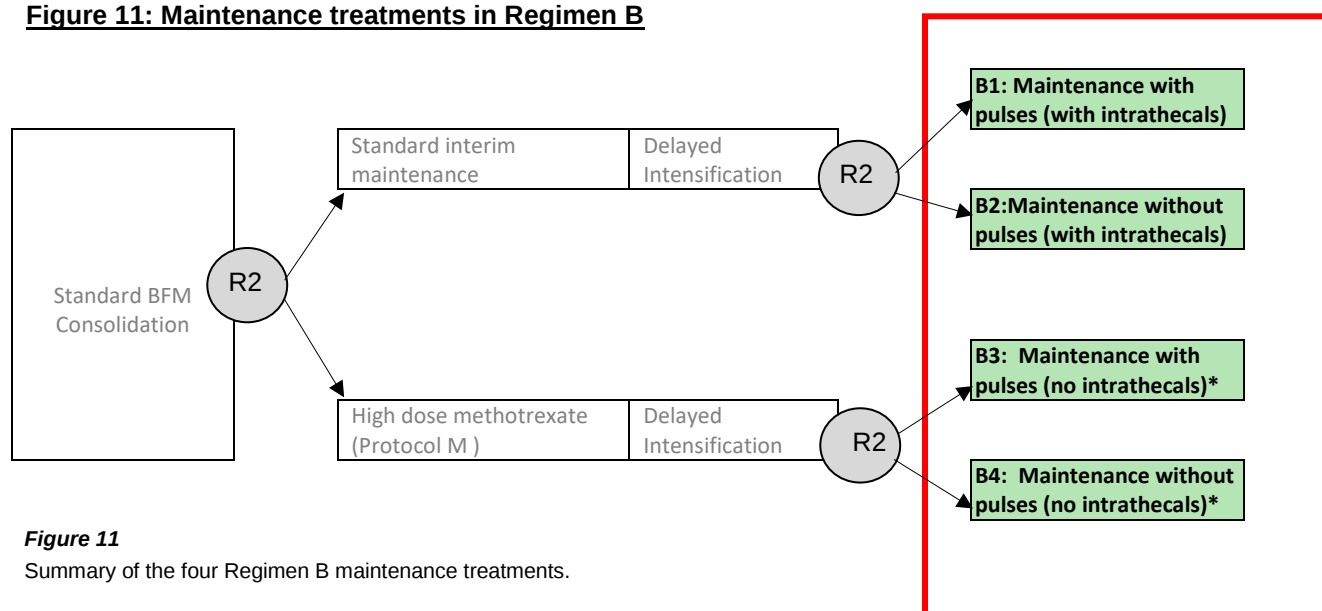


Figure 11

Summary of the four Regimen B maintenance treatments.

* T-ALL patients with a presenting WCC $>100 \times 10^9/L$ receive continuing intrathecal therapy.

7.6.8.1 Maintenance Regimen B1

This regimen is for patients randomised to receive standard interim maintenance and vincristine/dexamethasone pulses only.

This regimen is standard treatment for Regimen B patients and should be considered for those patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses on days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine for the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-Trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – ALL PATIENTS ONLY	Physiotherapy Assessment To be performed at weeks 31, 55 and 79 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life	QuEST and Quality of Life Questionnaire 4 must be completed

	questionnaire	at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment
--	---------------	---

7.6.8.2 Maintenance Regimen B2

This regimen is for patients randomised to receive standard interim maintenance and NO vincristine/dexamethasone pulses only.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
c)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
d)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 31, 55 and 79 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.6.8.3 Maintenance Regimen B3

This regimen is for patients randomised to receive high dose methotrexate (Protocol M) and vincristine/dexamethasone pulses only.

NB: Patients with T-ALL and a presenting WCC $>100 \times 10^9/L$ receive continuing intrathecal therapy.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

The IMPs in this treatment phase are highlighted in the table below.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses of days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate	20mg/m ² orally once per WEEK* Throughout maintenance. *Note that in T-ALL with presenting WCC $>100 \times 10^9/L$ none is given in the third week of each cycle for the first 6 cycles as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	T-ALL patients with presenting WCC $>100 \times 10^9/L$ ONLY On day 15 of each cycle, for the first 6 cycles only in T-ALL with presenting WCC $>100 \times 10^9/L$ Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine for the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.
TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC $>1.5 \times 10^9/L$. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – ALL PATIENTS ONLY	Physiotherapy Assessment To be performed at weeks 31, 55 and 79 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life	QuEST and Quality of Life Questionnaire 4 must be

	questionnaire	completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment
--	---------------	--

7.6.8.4 Maintenance Regimen B4

This regimen is for patients randomised to receive high dose methotrexate (Protocol M) and NO vincristine/dexamethasone pulses only.

NB: Patients with T-ALL and a presenting WCC $>100 \times 10^9/L$ receive continuing intrathecal therapy.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Doses Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per WEEK* throughout maintenance. *Note that in T-ALL with WCC $>100 \times 10^9/L$ none is given in the third week of each of the first 6 cycles as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
c)	Intrathecal methotrexate (THIS IS AN IMP)	T-ALL patients with presenting WCC $>100 \times 10^9/L$ ONLY On day 15 of each cycle, for the first 6 cycles only in T-ALL with presenting WCC $>100 \times 10^9/L$ Dose by age: <2yrs: 8mg 2yrs: 10mg >3yrs: 12mg. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
d)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC $>1.5 \times 10^9/L$. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – ALL PATIENTS ONLY	<u>Physiotherapy Assessment</u> To be performed at weeks 31, 55 and 79 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

Regimen C Treatment Schedule

7.6.9 Summary of Regimen C Treatment

TREATMENT PHASE	DURATION	DETAILS OF TREATMENT/RANDOMISATION
Registered patients Induction	Starts at day 15 (week 3). Continues to end of week 5.	It is recommended that patients registered on the study should be treated according to standard practice including dexamethasone 6mg/m ² /day orally for 28 days and standard MRD stratified post induction treatment.
Augmented BFM Consolidation	10 weeks (weeks 6-15)	
Capizzi Interim Maintenance	8 weeks (weeks 16-23)	Randomisation 2 (R2): Capizzi interim maintenance vs. high dose methotrexate + asparaginase (Protocol M-A)
Delayed Intensification	8 weeks (weeks 24-31)	
Maintenance	Continues for exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance in ALL and LBL.	Randomisation 2 (R2): vincristine and dexamethasone pulses vs. no pulses.

The following drugs require doses to be capped: dexamethasone in induction and vincristine.

7.6.10 Regimen C: Induction

It is recommended that patients registered on the study should be treated according to the standard arm with dexamethasone 6mg/m²/day orally for 28 days and standard MRD stratified post induction treatment.

This regimen is for patients with NCI Standard Risk BCP ALL who are subsequently found to have high risk cytogenetics, or Down syndrome patients with a slow early response.

NCI High Risk BCP ALL and all patients with T-ALL and high risk cytogenetics should receive Regimen B induction.

Patients switching to Regimen C induction must be before their 10th birthday at diagnosis and have a highest white cell count before starting treatment of <50x10⁹/L. In addition, Down Syndrome patients with a slow early response may also switch to Regimen C. Patients should switch from Regimen A at day 15 and should continue this induction modification for 21 days, regardless of the length of time that has elapsed since diagnosis.

Regimen B patients that are found to have high risk cytogenetics continue Regimen B induction and switch to Regimen C at consolidation.

This phase runs for 3 weeks from day 15 (beginning of week 3) to day 35 inclusive (end of week 5).

Patients with CNS disease at diagnosis

Patients with CNS disease at diagnosis should receive weekly intrathecal methotrexate until two consecutive clear CSFs have been obtained. See section 3.7.1 for details.

Use of intravenous dexamethasone in severely ill patients

Severely ill patients requiring intravenous dexamethasone at the beginning of Induction will not be excluded from randomisation. Intravenous dexamethasone should be given at the randomised dose and switched to oral therapy as soon as clinical circumstances allow.

a)	Dexamethasone	All patients should receive dexamethasone starting on day 1. Oral dexamethasone 6mg/m²/day (maximum dose 10mg/day in induction only) for a total of 28 days from commencing on regimen A and then tapered over the next 7 days i.e. to stop on day 35. The steroid should be divided into two doses per day. Down Syndrome patients aged >10 years: receive oral dexamethasone 10mg/m²/day for 7 days starting on day 15. The steroid should be divided into two doses per day. Do not cap dexamethasone dose.
b)	Vincristine	1.5mg/m ² (maximum single dose 2mg) intravenous weekly on days 16, 23 and 30
c)	Daunorubicin	45mg/m ² intravenous over 1 hour (or as a bolus alongside a fast running infusion in adults) on days 16 and 23. Note that this dose

		is higher than that in regimen B and is for two doses only.																				
d)	Pegaspargase (Oncaspar)	1000 iu/m ² intramuscular on day 18.																				
e)	Intrathecal methotrexate	On day 29. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. NB Patients who have CNS disease at presentation should receive weekly doses until two clear CSF samples are obtained (see section 3.7.1). Do not schedule vincristine on the same day as the intrathecal methotrexate																				
f)	Mercaptopurine	60mg/m ² /day orally once a day starting on day 29 (beginning week 5) (if neutrophils >0.75x10 ⁹ /L and platelets >75x10 ⁹ /L) and continuing to day 14 of consolidation (3 weeks from the start in week 5 of induction). If necessary give extra doses between induction and consolidation to ensure continuity of therapy. Do not adjust dose according to blood count. NB: the mercaptopurine dose in Regimen C induction is different to that given in Regimen A.																				
g)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	This drug is given as PCP prophylaxis orally twice a day (bd) on 2 consecutive days each week starting from day 1. Dose for children is based on surface area as detailed below: <table><tr><th>Surface area</th><th>Co-Trimoxazole</th><th>Trimethoprim</th><th>Sulphamethoxazole</th></tr><tr><td>0.5-0.75m²</td><td>240mg bd</td><td>40mg bd</td><td>200mg bd</td></tr><tr><td>0.76-1.0m²</td><td>360mg bd</td><td>60mg bd</td><td>300mg bd</td></tr><tr><td>1.1m² – 1.5m²</td><td>480mg bd</td><td>80mg bd</td><td>400mg bd</td></tr><tr><td>*Over 1.5 m²</td><td>480-960mg</td><td>80-120mg bd</td><td>400-800mg bd</td></tr></table> *according to local policy over 1.5m² For adults, the recommended PCP prophylaxis is Co-trimoxazole 960mg bd for 2 consecutive days each week. See also section 7.11.3 for details of alternative PCP prophylaxis regimens and permitted dose modifications for toxicity.	Surface area	Co-Trimoxazole	Trimethoprim	Sulphamethoxazole	0.5-0.75m ²	240mg bd	40mg bd	200mg bd	0.76-1.0m ²	360mg bd	60mg bd	300mg bd	1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd	*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd
Surface area	Co-Trimoxazole	Trimethoprim	Sulphamethoxazole																			
0.5-0.75m ²	240mg bd	40mg bd	200mg bd																			
0.76-1.0m ²	360mg bd	60mg bd	300mg bd																			
1.1m ² – 1.5m ²	480mg bd	80mg bd	400mg bd																			
*Over 1.5 m ²	480-960mg	80-120mg bd	400-800mg bd																			

TESTS	Bone Marrow (mandatory) - <u>ALL PATIENTS ONLY</u>	Bone marrow should be checked at day 29. Send day 29 marrow for MRD.
	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood in EDTA tube between Days 16 (12-18*) & 30 (25-31) * If sent on day 18 ensure sample is taken before the day 18 asparaginase. See Appendix 12 for details of the samples required.
	Closed to recruitment 11-Jan-2018	<u>Patients randomised to short dexamethasone (with split dosing on days 1-7 and 15-21)</u> Day 21: blood samples at 1, 2, 4 and 8 hours after the first dose of

	Dexamethasone Study samples (optional – consent must have been provided) - <u>ALL PATIENTS ONLY</u>	dexamethasone on day 28. <u>Patients randomised to standard dexamethasone</u> Day 28: blood samples at 1, 2, 4 and 8 hours after the first dose of dexamethasone on day 28. Appendix 15 – Dexamethasone pharmacokinetic study - CLOSED for details of the samples required
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Samples</u> Blood samples taken before and after the first vincristine dose on day 2. Saliva sample taken for genetic analysis prior to vincristine PK sampling. <u>Physiotherapy Assessment</u> To be performed at diagnosis. See Appendix 24 for details of the samples required.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 1 should already have been completed (after informed consent has been obtained ideally prior to randomisation). Must be completed by end of Week 1 at the latest. QuEST and Quality of Life Questionnaire 2 must be completed at the end of induction and within 2 weeks of this time point at the latest.

7.6.11 Regimen C: Post-induction therapy

Acute Lymphoblastic Leukaemia

This regimen is for the following patients:

- 1) Patients with MRD risk status at day 29
- 2) Patients with **high-risk cytogenetics** (see section 3.3.1) regardless of day 29 MRD
- 3) Patients in the day 29 MRD NO Result Group who showed a slow early response to induction therapy at day 8 (receive Regimen B induction) or 15 (receive Regimen A induction)
- 4) Patients with partial response BMA M2 (5-25% blasts at day 29). See section 3.7.5.2.
- 5) Patients with persisting CNS disease at day 29, regardless of MRD risk.
- 6) Down syndrome ALL patients whose day 15 bone marrow showed a slow early response. See Appendix 5 for further guidance on treatment.

All the above patients should receive Regimen C consolidation and MRD should then be measured again on recovery of blood counts after week 14 of chemotherapy.

Patients with MRD $\geq 0.5\%$ on recovery from consolidation are deemed **MRD High Risk** and are not eligible for R2

Patients with MRD $< 0.5\%$ on recovery from consolidation at week 14 are deemed **MRD Intermediate Risk** and should continue Regimen C with a single DI.

Patients in the **MRD NO result** group are eligible for the factorial methotrexate and pulses randomisation (R2) and should be randomised after day 29. After 14 weeks of chemotherapy they should continue Regimen C. No further MRD assessment is required in these patients.

MRD Intermediate Risk patients are eligible for the factorial methotrexate and pulses randomisation which should be performed as soon as possible after receipt of the MRD result upon recovery from consolidation at week 14.

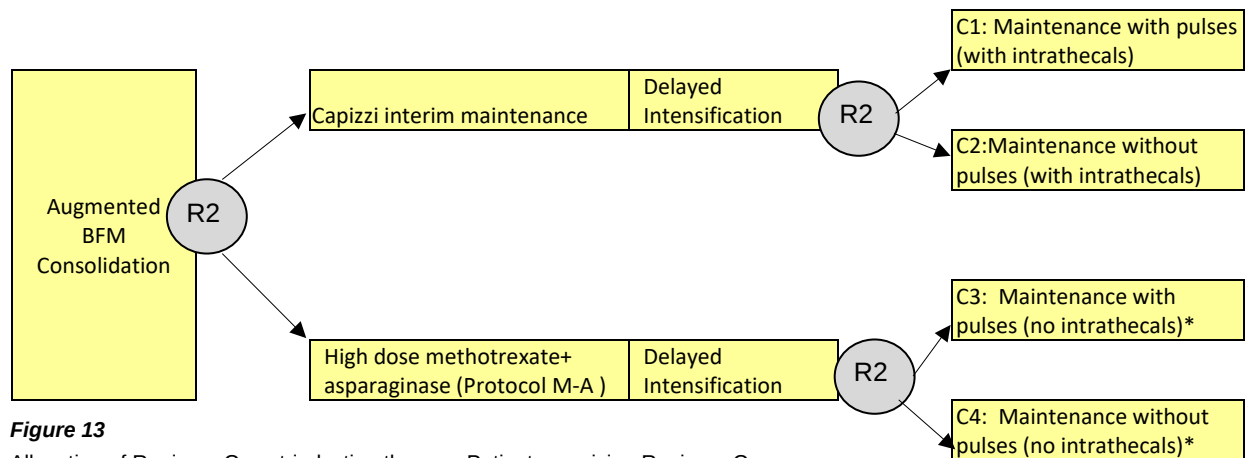
Lymphoblastic Lymphoma

This regimen is intended for the following LBL patients with a good response to induction “good responders” (see section 3.5):

- 1) LBL patients with T-cell immunophenotype
- 2) B-cell precursor LBL patients with high risk cytogenetics

These patients are eligible for the factorial methotrexate and pulses randomisation which should be performed as soon as possible after receiving the tumour volume assessment result.

Patients with $< 35\%$ reduction in tumour volume after induction are taken off protocol treatment.

Figure 13: Allocation of post-induction treatment in Regimen C**Figure 13**

Allocation of Regimen C post-induction therapy. Patients receiving Regimen C are randomised (R2) into four groups to receive standard interim maintenance or high dose methotrexate + asparaginase (Protocol M-A) followed by a single delayed intensification and either maintenance therapy with pulses or maintenance therapy without pulses

* T-ALL patients with a presenting WCC $>100 \times 10^9/L$ receive continuing intrathecal therapy.

7.6.12 Regimen C: Augmented BFM Consolidation

This phase runs for 10 weeks from day 1 (beginning of week 6) to day 70 inclusive (end of week 15).

Patients should have an absolute neutrophil count (ANC) $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase. If blood counts had recovered to ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ at week 5 but subsequently fallen again, continue with week 6 therapy as long as patient clinically well.

Once consolidation has begun it can be interrupted for myelosuppression (ANC $<0.75 \times 10^9/L$ or platelets $<75 \times 10^9/L$) at day 29, but once the cyclophosphamide has been given at day 1 or 29, therapy should continue, except in patients who are febrile **and proven to have infection** (i.e. do not stop because of neutropenia alone). Treatment should restart when signs of infection have abated.

Upon recovery from consolidation, a bone marrow sample should be taken for patients with ALL. Upon count recovery (ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$), week 15 should begin - all Regimen C patients receive a week of holding chemotherapy with mercaptopurine at $25\text{mg}/\text{m}^2/\text{day}$ from day 64-70 (i.e. week 15) whilst awaiting the post-consolidation MRD result.

NB the mercaptopurine dose at week 15 is lower than the dose given earlier in this phase.

Co-Trimoxazole should be omitted during week 15 pending the R2 randomisation allocation.

There are no IMPs in this treatment phase.

a)	Cyclophosphamide	1000mg/m ² intravenous over 20-30 minutes on days 1 and 29. Give 125mls/m ² /hr of Dextrose /Saline infusion for 30 minutes before the cyclophosphamide and for 3.5 hours afterwards, i.e. 4 hours in total. Mesna is not needed.
b)	Cytarabine	75mg/m ² intravenous or subcutaneously. Give on 4 consecutive days in weeks 6, 7, 10 and 11 starting on the same day each week. Cytarabine pulses should ideally start the day after cyclophosphamide when both drugs are given in the same week.
c)	Mercaptopurine	All patients receive oral mercaptopurine $60\text{mg}/\text{m}^2/\text{day}$ once a day for 21 days, beginning week 5 of induction to end of week 2 of consolidation, and again for 14 days on days 29-42 of consolidation. Do not adjust dose according to blood count. A 'holding chemotherapy' dose of $25\text{mg}/\text{m}^2/\text{day}$ should be given once a day on days 64-70 (week 15). This is count dependent. If necessary give extra doses between consolidation and interim maintenance/Protocol M-A to ensure continuity of therapy.
d)	Vincristine	1.5mg/m ² intravenous (maximum single dose 2mg) on day 16, 23, 44 and 51
e)	Pegaspargase (Oncaspar)	1000 units/m ² intramuscular on days 16 and 44

f)	Intrathecal methotrexate	On days 1, 8 and 22. Dose by age: <2 yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg Do not schedule vincristine on the same day as intrathecal methotrexate.
g)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section. Omit for all patients during week 15.

TESTS	Bone Marrow - <u>ALL PATIENTS ONLY</u>	Bone marrow should be checked upon count recovery from consolidation at week 14 <u>An additional bone marrow sample is required at day 22 (Week 9) for Regimen C (ALL patients only).</u> This will be used in a prospective blinded analysis of MRD clearance in order to improve understanding of MRD kinetics in this group.
	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood EDTA tube on any ONE occasion between days 24-30 <u>or</u> days 52-58 See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed during weeks 6-8, following administration of the first 5 vincristine doses.

7.6.13 Regimen C: Capizzi Interim Maintenance

This regimen is for patients randomised to Capizzi interim maintenance only.

This regimen is standard treatment for Regimen C patients and should be considered for those patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

This phase runs for 8 weeks from day 1 (beginning of week 16), to day 56 inclusive (end of week 23).

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase.

During Capizzi interim maintenance, therapy should be interrupted for serious infection such as Varicella or pneumocystis (presumed or proven). The last IMP will be administered on day 42, regardless of the number of methotrexate doses that have been omitted. The timings of administration of the drugs are important; do not reschedule any of the components. Approximately 10-15% of patients are able to escalate the methotrexate dose to the maximum of $300\text{mg}/\text{m}^2$. About 30-40% are able to make some escalation. The major toxicity is likely to be mucositis. The full toxicity grading is included in the section on toxicity assessment and reporting.

Start Capizzi at a methotrexate dose of $100\text{mg}/\text{m}^2$. Increase the dose for each subsequent dose by $50\text{mg}/\text{m}^2$, as tolerated, to a maximum dose of $300\text{mg}/\text{m}^2$. Prior to each dose patient should be assessed for 1) oral, 2) haematological, 3) hepatic and 4) renal toxicity, and the dosage modified as outlined below. Doses of vincristine and Pegaspargase should only be omitted for serious intercurrent illness.

- 1) Oral toxicity: for grade 2 mucositis of over 3 days duration, decrease intravenous MTX dose by 30% to 70% of previous dose then escalate each subsequent dose by $50\text{mg}/\text{m}^2$, as tolerated. For grade 3-4, mucositis, withhold intravenous MTX until resolved; resume at 50% of the previously attained dose and subsequently escalate to 75% then to 100% dose at 10 day intervals provided grade 3-4 toxicity does not recur. Consider culturing lesions for herpes simplex if mucositis persists or recurs.
- 2) Haematological toxicity: omit methotrexate if $\text{ANC} < 0.75 \times 10^9/L$ or platelets $< 75 \times 10^9/L$; it should be reinstituted on the first due date following the omitted dose when $\text{ANC} > 0.75 \times 10^9/L$ and platelets $> 75 \times 10^9/L$, and the dosage should be decreased to 80% of the previously administered dose. Escalate each subsequent dose by $50\text{mg}/\text{m}^2$, as tolerated. Missed doses will not be made up. If counts do not recover within 21 days, check bone marrow status.
- 3) Liver Dysfunction: If bilirubin is $> 50 \mu\text{mol}/L$ omit intravenous MTX until it is less than $20 \mu\text{mol}/L$, and then restart at 50% of the previously attained dose. Escalate from 50% to 75% to 100% dose at 10-day intervals provided hyperbilirubinaemia does not recur. Do not modify dosage for elevated aminotransferases.
- 4) Kidney Dysfunction (Grade 3-4): Omit intravenous MTX until grade 0 toxicity (i.e. completely resolved). Resume at 100% of the previously attained dose and then escalate each subsequent dose by $50\text{mg}/\text{m}^2$, as tolerated and continue at 10-day intervals.

The IMPs in this treatment phase are highlighted in the table below.

a)	Vincristine (THIS IS AN IMP)	1.5mg/m ² intravenous (maximum single dose 2mg) as a single dose on days 2, 12, 22, 32, and 42.
b)	Methotrexate (THIS IS AN IMP)	100mg/m ² intravenous over 10-15 minutes as initial dose on day 2. Escalate subsequent doses by 50mg/m ² to toxicity and modify dosage as necessary according to the above guidelines on days 12, 22, 32 and 42
c)	Pegaspargase (Oncaspar) (THIS IS AN IMP)	1000 units/m ² intramuscular on day 3 and day 23.
d)	Intrathecal methotrexate (THIS IS AN IMP)	On day 1 and 31 (or as close as possible). Dose by age: <2 yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg Do not schedule vincristine on the same day as intrathecal methotrexate.
e)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood in EDTA tube on day 32 (31-37). See Appendix 12 for details of the samples required.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.

7.6.14 Regimen C: High dose methotrexate + asparaginase (Protocol M-A)

This regimen is intended for patients randomised to high dose methotrexate and asparaginase (Protocol M-A).

The administration of high dose methotrexate (HDMTX) carries significant potential toxicity which in extreme cases can be fatal. This is prevented by effective folinic acid rescue. Absolute compliance with the schedule of hydration, monitoring of levels and folinic acid rescue is mandatory in this phase of the protocol. It is vital to read this section of the protocol and Appendix 7 fully **before** commencing administration of HDMTX. Please also ensure that you have made certain that your patient has no contraindication to HDMTX as detailed in section 0. **Please note that to be eligible for entry into this phase renal function should be within normal limits corrected for age.**

This phase runs for 8 weeks from day 1 (beginning of week 16) to day 56 inclusive (end of week 23) (i.e. 8 weeks). **NB the first high dose methotrexate for Regimen C patients is given on day 1 of Protocol M-A which is different to Protocol M in Regimens A and B.**

Prior to starting the first pulse of HDMTX patients must be free of infection, diarrhoea and mucositis and should have an ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$ to start this phase. Renal function should be within normal limits corrected for age

Prior to starting each subsequent pulse of HDMTX serum creatinine must be $<1.5 \times$ baseline or GRF creatinine clearance $>65\text{mL/minute}/1.73\text{m}^2$. If renal function does not recover, omit MTX. Do not give HDMTX to a patient with this degree of renal impairment, assuming that prolonged excretion can be managed with Carboxypeptidase. Counts permitting, continue with mercaptopurine regardless of renal function throughout protocol M/M-A to 2 weeks after last course of HD MTX. ALT and AST should be less than 5 x normal. Wherever possible avoid concurrent NSAIDs, aminoglycosides and other nephrotoxic drugs. See Appendix 6 for information on drug interactions.

Prior to starting each subsequent pulse of high dose methotrexate ANC should be $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$. If blood counts are not adequate on the scheduled day of infusion then stop 6-MP. On recovery to ANC $>0.5 \times 10^9/L$ and platelets $>50 \times 10^9/L$ then administer the scheduled dose of methotrexate and restart 6-MP at full 25mg/m^2 dose.

The IMPs in this treatment phase of highlighted in the table below.

a)	Mercaptopurine (THIS IS AN IMP)	$25\text{mg/m}^2/\text{day}$ orally once a day on days 1-49 (weeks 16-22).
b)	Methotrexate (THIS IS AN IMP)	5 g/m^2 intravenous on day 1, 15, 29 and 43. All patients must be prehydrated as per guideline in Appendix 7. The initial 10% of dose is delivered over 30 minutes, the remaining 90% over 23 ½ hours. NB method and schedule of administration should be guided by Appendix 7. Timings of drug administration must be followed strictly.
c)	Folinic acid	15mg/m^2 intravenously 42, 48 and 54 hours after the start of each methotrexate infusion. Further details of the folinic acid rescue are given in Appendix 7. Timings of drug administration must be followed strictly.
d)	Intrathecal methotrexate (THIS IS AN IMP) Intrathecal	Should optimally be given within 2 hours after the start of methotrexate infusion on day 1, 15, 29 and 43. Dose by age:

	methotrexate contd. (THIS IS AN IMP)	<2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg
e)	Pegaspargase (THIS IS AN IMP)	1000 units/m ² intramuscularly on days 3 and 23.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Must be omitted during Protocol M-A (Re-start after day 46 when folinic acid rescue is complete.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood EDTA on day 13 (11- 17) See Appendix 12 for details of the samples required.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 3 must be completed at the end of interim maintenance and within 2 weeks of this time point at the latest.

7.6.15 Regimen C: Delayed Intensification

This phase runs for 8 weeks from day 1 (beginning of week 24) to day 56 (end of week 31).

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$ to start this phase.

This phase consists of reinduction and reconsolidation. Once started, reinduction and reconsolidation are not interrupted for myelosuppression alone.

Reconsolidation is scheduled to begin on day 29, but should be delayed until ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$.

Treatment may be interrupted for **serious** infection (presumed or proven) such as Varicella, pneumocystis pneumonia, or neutropenia with fever.

Bone marrows, which may be indicated because of persistent cytopenias, are often difficult to interpret in this phase. Pancytopenia is inevitable and M2 recovery marrows are common. **Note that delayed intensification in regimen C includes pegaspargase and additional doses of vincristine in the reconsolidation phase.**

There are no IMPs in this treatment phase.

Reinduction (weeks 24-27)		
a)	Vincristine	1.5mg/m ² intravenous (maximum single dose 2mg) on days 2, 9 and 16.
b)	Doxorubicin	25mg/m ² intravenous over 1 hour (or as a bolus alongside a fast running infusion in adults) on days 2, 9 and 16.
c)	Dexamethasone	10mg/m ² /day orally divided into two doses on days 2-8 and 16-22. Do not cap dexamethasone dose.
d)	Intrathecal methotrexate	On day 1. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine on the same day as intrathecal methotrexate
e)	Pegaspargase (Oncaspar)	1000 units/m ² intramuscular on day 4
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be given throughout this phase of treatment as per guidelines provided in induction regimen section.
Reconsolidation (weeks 28-31)		
Patients should have an ANC $>0.75 \times 10^9/L$ and platelets of $>75 \times 10^9/L$ to start this phase		
g)	Cyclophosphamide	1000mg/m ² intravenous over 20-30 minutes on day 29. Give 125mls/m ² /hr of Dextrose /Saline infusion for 30 minutes before the cyclophosphamide and for 3.5 hours afterwards, i.e. 4 hours in total. Mesna is not needed
h)	Mercaptopurine	All patients receive mercaptopurine 60mg/m²/day during consolidation. The drug is given once a day by mouth for 14 days from day 29 (beginning of week 28) to day 42 (end of week 29). No dose adjustments are made during this block.
i)	Cytarabine	75mg/m ² intravenous or subcutaneously. Give on 4 consecutive

		days in weeks 28 and 29 starting on the same day each week. Cytarabine pulses should ideally start the day after cyclophosphamide when both drugs are given in the same week.
j)	Vincristine	1.5mg/m ² intravenous on days 43 (week 30) and 50 (week 31).
k)	Pegaspargase (Oncaspar)	1000 units/m ² intramuscular on day 43 (week 30).
l)	Intrathecal methotrexate	On days 29 and 36. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12 mg.
m)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Peripheral blood EDTA tube Day 16 (12-18). See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed during weeks 28-31 of delayed intensification and before the onset of maintenance.

7.6.16 Regimen C: Maintenance

Note regarding randomisations. As per Figure 14 four maintenance regimens will be employed in UKALL 2011. All four employ daily oral mercaptopurine and weekly oral methotrexate. **Patients randomised to receive high dose methotrexate in interim maintenance do not receive continuing intrathecal therapy except in patients with T-ALL and a presenting WCC >100 $\times 10^9/L$.** Patients randomised to receive “No pulses” do not receive monthly pulses of vincristine and dexamethasone.

Maintenance runs from day 1 cycle 1 (beginning of week 32). The duration of each cycle is 12 weeks. Treatment is stopped exactly 2 years (for girls) or 3 years (for boys) from the start of interim maintenance/Protocol M-A for both ALL and LBL. The cycle in progress is stopped when this date is reached. No allowances are made for interruptions in treatment after the start of interim maintenance.

Patients should have an ANC $>0.75 \times 10^9/L$ and platelets $>75 \times 10^9/L$ to start this phase. Once maintenance is started the clock does not stop regardless of whether treatment is given or not. Only mercaptopurine and oral methotrexate will be interrupted for myelosuppression and the time is not made up. Vincristine and dexamethasone pulses (if given) should be given regardless of blood count. Days off therapy for intercurrent infections are counted as days of maintenance and the time is not made up.

Anaemia occurring in the course of maintenance therapy should be treated with transfusion and the dose of drug is maintained. If *persistent anaemia* occurs (i.e., haemoglobin below 8 g/dl) investigate for Parvovirus infection. Please contact the Chief Investigator or Co-investigators for advice.

Figure 14: Maintenance treatments in Regimen C

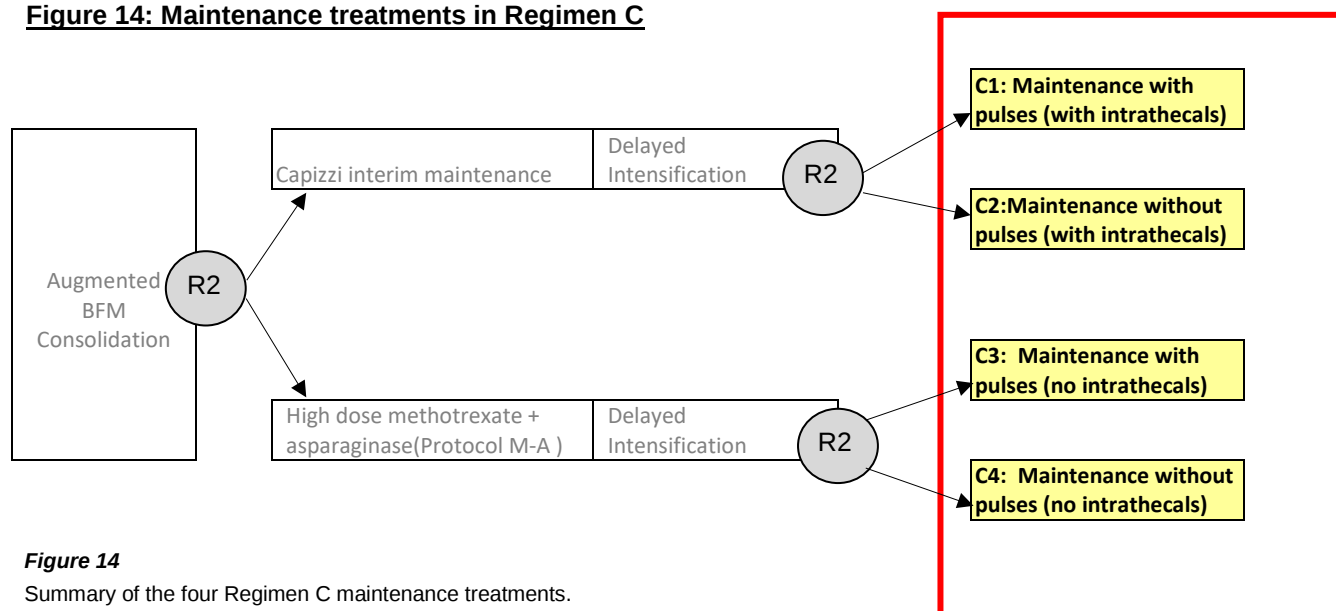


Figure 14

Summary of the four Regimen C maintenance treatments.

7.6.16.1 Maintenance Regimen C1

This regimen is for patients randomised to receive standard Capizzi interim maintenance and vincristine/dexamethasone pulses only.

This regimen is standard treatment for Regimen C patients and should be considered for patients who are not randomised due to refusal, toxicity or specific exclusion from high dose methotrexate.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses on days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle (day 15) as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine for the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction regimen section.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 36, 60 and 84 of maintenance. To be performed at the end of treatment.

QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment
-----	-------------------------------	---

7.6.16.2 Maintenance Regimen C2

This regimen is for patients randomised to receive standard Capizzi interim maintenance and NO vincristine/dexamethasone pulses only.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per week during weeks beginning days 1, 8, 22, 29, 36, 43, 50, 57, 64, 71 and 78 of each cycle. Note none is given in the third week of each cycle as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
c)	Intrathecal methotrexate (THIS IS AN IMP)	On day 15 of each cycle. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs:12mg. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
d)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction treatment regimen.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 36, 60 and 84 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.6.16.3 Maintenance Regimen C3

This regimen is for patients randomised to receive high dose methotrexate + asparaginase (Protocol M-A) and vincristine/dexamethasone pulses only.

Please note that in T-ALL with WCC $>100 \times 10^9/L$ intrathecal methotrexate is given in the third week of each of the first 6 cycles. No oral methotrexate is given during those weeks.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Dexamethasone (THIS IS AN IMP)	6mg/m ² /day orally, divided into twice-daily doses on days 1-5, 29-33 and 57-61 of each cycle.
b)	Vincristine (THIS IS AN IMP)	1.5mg/m ² (maximum single dose 2mg) intravenous on days 1, 29, and 57 of each cycle.
c)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
d)	Oral methotrexate	20mg/m ² orally once per WEEK* throughout maintenance. *Note that in T-ALL with WCC $>100 \times 10^9/L$ none is given in the third week of each of the first 6 cycles as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
e)	Intrathecal methotrexate (THIS IS AN IMP)	T-ALL patients with presenting WCC $>100 \times 10^9/L$ ONLY On day 15 of each cycle, for the first 6 cycles only in T-ALL with presenting WCC $>100 \times 10^9/L$. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Do not schedule vincristine for the same day as intrathecal methotrexate. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
f)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction treatment regimen.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC $>1.5 \times 10^9/L$. See Appendix 12 for details of the samples required.
--------------	--	--

	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 36, 60 and 84 of maintenance. To be performed at the end of treatment.
QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months Quest and Quality of Life Questionnaire 5 must be completed at the end of treatment

7.6.16.4 Maintenance Regimen C4

This regimen is for patients randomised to receive high dose methotrexate + asparaginase (Protocol M-A) and NO vincristine/dexamethasone pulses only.

Please note that in T-ALL with WCC $>100 \times 10^9/L$ intrathecal methotrexate is given in the third week of each of the first 6 cycles. No oral methotrexate is given during those weeks.

The IMPs in this treatment phase are highlighted in the table below.

NB. Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

a)	Mercaptopurine	75mg/m ² /day orally once a day throughout maintenance. Dose adjustments are described in section 7.11.2.
b)	Oral methotrexate	20mg/m ² orally once per WEEK* throughout maintenance. *Note that in T-ALL with WCC $>100 \times 10^9/L$ none is given in the third week of each of the first 6 cycles as an intrathecal dose is given during that week. Dose adjustments are described in section 7.11.2.
c)	Intrathecal methotrexate (THIS IS AN IMP)	T-ALL patients with presenting WCC $>100 \times 10^9/L$ ONLY On day 15 of each cycle, for the first 6 cycles only in T-ALL with presenting WCC $>100 \times 10^9/L$. Dose by age: <2yrs: 8mg 2yrs: 10mg ≥3yrs: 12mg. Oral methotrexate is omitted during the weeks intrathecal methotrexate is given.
d)	Co-trimoxazole (trimethoprim and sulphamethoxazole)	Should be continued throughout this phase of treatment as per guidelines provided in induction treatment regimen.

TESTS	Closed to recruitment 25-Jul-2017 Asparaginase Study samples (optional – consent must have been provided)	Sample should be taken at any convenient point during maintenance. Please ensure that WCC $>1.5 \times 10^9/L$. See Appendix 12 for details of the samples required.
	Closed to recruitment 31-Mar-2018 Vincristine Study (optional – consent must have been provided) – <u>ALL PATIENTS ONLY</u>	<u>Physiotherapy Assessment</u> To be performed at weeks 36, 60 and 84 of maintenance. To be performed at the end of treatment.

QoL	Quality of Life questionnaire	QuEST and Quality of Life Questionnaire 4 must be completed at 18 months QuEST and Quality of Life Questionnaire 5 must be completed at the end of treatment
-----	-------------------------------	---

7.7 Assessments

Centres treating patients with leukaemia and lymphoma are expected to have appropriate investigation protocols for the pre-treatment assessment of new patients, for monitoring progress during treatment and once therapy has ended, and for dealing with any complications that may arise.

7.7.1 Routine Clinical and Laboratory Assessments during treatment

- o Clinical examinations will be carried out as part of routine clinical care.
- o Assessment of performance status (Lansky or ECOG) will be carried out before each discrete block of therapy.
- o Height/weight & BSA will be assessed as needed to prescribe each block of therapy
- o Full blood count/and other laboratory tests e.g. LFTs, U&Es will be carried out at the clinicians discretion as part of the routine management of acute lymphoblastic leukaemia and lymphoblastic lymphoma. These tests are usually carried out a minimum of three times weekly during inpatient stays.

7.8 Sample Collection

This section provides information about the samples/tests required throughout the trial. A summary schedule of events can also be found on page 27 for ALL patients and page 28 for LBL patients.

NB. If any mandatory bone marrow tests have been omitted on the first diagnostic sample, a second sample must be taken before treatment starts. A trephine biopsy is required if an adequate sample cannot be obtained by aspiration.

In patients in whom clinical condition precludes bone marrow aspirate then mandatory tests can be performed on peripheral blood.

Patients registered in the study but NOT participating in the trial randomisation should only have tests performed that are part of normal clinical practice.

Informed consent for optional sub-studies must be obtained before any additional tests are performed.

7.8.1 Diagnosis

Test	Details	Optional sub-study?
Blood and/or bone marrow for morphology review	Blood and bone marrow for morphology assessment locally.	No
Bone marrow for MRD test - <u>ALL PATIENTS ONLY</u>	Bone marrow aspirate for MRD measurement to be sent to a UK MRD network laboratory. 2-5ml of bone marrow from all patients with suspected leukaemia should be placed into an ACD container supplied by the local MRD network laboratory. Do NOT use EDTA or other transport media. In patients with peripheral blasts counts greater than $20 \times 10^9/L$, 5ml of blood collected into ACD is also acceptable at diagnosis. Please note that peripheral blood is of no value as an MRD sample at other time points.	No

	See section 7.9 for details on sample transport and processing.	
Blood and/or bone marrow for diagnostic flow cytometry	Flow cytometric diagnosis of ALL should be made according to local practice within a CPA approved laboratory. Recommended diagnostic antibody panels are specified in Appendix 23. NB. in T-ALL a mandatory panel of antibodies is required in order to identify ETP ALL.	No
Bone marrow for morphology review	Bone marrow for morphology assessment locally. Required in case of MRD failure.	No
Bone marrow for cytogenetics	Send bone marrow sample to regional genetic/cytogenetic laboratory according to local arrangements. Do NOT use EDTA transport media. If necessary, contact local laboratory for transport details. See 5.1 for details.	No
Closed to recruitment 25-Jul-2017 Asparaginase Study samples	5ml of bone marrow aspirate collected from a separate aspirate site to the MRD sample in ACD-A tube 5ml of peripheral blood in EDTA tube See Appendix 12 for further details.	Yes

7.8.1.1 Collection of diagnostic samples from LBL patients at diagnosis

In the management of a patient presenting with a mediastinal mass then immediate patient safety takes priority over establishment of a histological diagnosis. This is discussed further in Appendix 22. Expert anaesthetic opinion will guide the timing of any diagnostic procedure. Prior cytoreductive therapy that makes anaesthesia safe might be necessary prior to any diagnostic attempt. Procedures that might allow a diagnosis to be made without the need for general anaesthesia might be considered.

When a diagnostic procedure is safely attempted samples should be taken for histopathology including immunophenotyping in the first instance and then for cytogenetic analysis and for tumour storage for future research studies (cell banking). Under the same anaesthetic staging investigations including bone marrow and lumbar puncture should be undertaken.

7.8.2 During induction

Time point	Test	Reg. A	Reg. B	Reg. C	Details	Optional Sub-study?
Day 2	Vincristine Study – <u>ALL PATIENTS ONLY</u>	✓	✓	✓	Blood samples taken before and after the first vincristine dose on day 2. Saliva sample taken for genetic analysis prior to vincristine PK sampling. See Appendix 24 for details	Yes
Day 8	Bone marrow for morphology review - <u>ALL PATIENTS ONLY</u>	✓	✓		Bone marrow for morphology assessment locally. Required in case of MRD failure.	No
Day 15	Bone marrow for morphology review - <u>ALL PATIENTS ONLY</u>	✓			Test performed locally. Required in case of MRD failure.	No
Day 29	Bone marrow for MRD test - <u>ALL PATIENTS ONLY</u>	✓	✓	✓	5mls of bone marrow should be placed into ACD (and only as a last resort EDTA). Peripheral blood is NOT an acceptable alternative. See section 7.9 for details on sample transport and processing	No

7.8.3 During Consolidation

Time point	Test	Reg. A	Reg. B	Reg. C	Details	Optional Sub-study?
Consolidation (week 9)	Bone marrow for MRD test - <u>ALL PATIENTS ONLY</u>			✓	5mls of bone marrow should be placed into ACD (and only as a last resort EDTA). Peripheral blood is NOT an acceptable alternative. See section 7.9 for details on sample transport and processing	No
Upon recovery from consolidation (week 14)	Bone marrow for MRD test - <u>ALL PATIENTS ONLY</u>			✓	5mls of bone marrow should be placed into ACD (and only as a last resort EDTA). Peripheral blood is NOT an acceptable alternative. See section 7.9 for details on sample transport and processing	No

7.8.4 During Maintenance

Time point	Test	Reg . A	Reg . B	Reg . C	Details	Optional Sub-study?
Any convenient time during maintenance	Asparaginase Study samples	✓	✓	✓	10ml of peripheral blood in EDTA tube at any convenient time point during maintenance. Please ensure that WCC >1.5 x 10 ⁹ /L. See Appendix 12 for further details.	Yes

7.9 MRD sample requirements

The tables in section 7.8 detail what is required for MRD analysis at the various time points in this trial. See also Appendix 4 for MRD laboratory details and instructions on sample collections and transport to the MRD laboratory.

Request forms

These will be supplied with the ACD bottles. They have been designed to provide sufficient information for each patient to be reliably identified whilst at the same time attempting to blind the network lab to clinical risk. Thus whilst the date of birth is required for identification and in due course the immunophenotype for directed PCR screening, we will not require information about WCC or treatment arm. We do wish to know if the patient has Philadelphia positive ALL so we can make RNA for MRD analysis.

Transport

MRD samples should be sent to your local MRD laboratory using either Royal Mail Safe boxes or by courier. Accounts with couriers are not centrally managed and if delivery by courier is the preferred route this should be arranged with your local MRD laboratory. Royal Mail Safe boxes can be obtained from MRD laboratories. Appendix 4 provides a list of MRD laboratories and their catchment Principal Treatment Centres, and the contact details for the UK MRD network laboratories.

Sample processing

On receipt of bone marrow aspirates, the MRD laboratory will assign the patient and the sample a unique number according to the standard operating procedure. Cell counts will be recorded and DNA extracted within 24 hours of receipt of the sample. A minimum of 10 micrograms of DNA is required at diagnosis and 5 micrograms at end of induction. In the event that an inadequate sample is obtained then a further sample will be requested.

7.10 Quality of Life questionnaires

Quality of life assessment (including QuEST) is a mandatory for all patients participating in Registration and /or R2 of the UKALL 2011 trial. This is assessed for all patients using a questionnaire at the following time points:

1. **During week 1.** Baseline assessment (No burden of care questionnaire at this time point)
2. **End of induction treatment.** This allows a direct comparison of the induction regimens.
3. **At the end of interim maintenance.** This allows comparison of the immediate impact of intravenous methotrexate.
4. **At 18 months.** This allows comparison of the impact of therapy on patients who have previously received methotrexate and those who are receiving pulses of dexamethasone and vincristine.
5. **At the end of treatment.** A final assessment at the completion of therapy. This allows evaluation of the length of treatment on QoL and family burden of care, with ongoing comparison between the groups randomised to pulses of vincristine and dexamethasone and those not receiving these.

See appendix 13 for full details on the Quality of Life Study.

7.11 Dose Modifications

7.11.1 Dexamethasone

Where there is **clinical urgency only** (e.g. hyperleucocytosis, mediastinal mass) treatment should not be delayed and dexamethasone may be initiated at standard dose (6mg/m² per day) before trial entry

7.11.2 Mercaptopurine (6-MP) and methotrexate (MTX)

Only 6-MP and MTX will be interrupted for myelosuppression in interim or continuing maintenance or Regimen A consolidation. The omitted doses will not be made up.

TPMT genotype and mercaptopurine (6-MP) dose adjustment (see Appendix 21 for details)

The thiopurine drug 6-MP is central to successful therapy of lymphoid malignancy in children. The cytotoxic effect of 6-MP is exerted by drug-derived thioguanine nucleotides (TGNs). The rate of TGN formation from 6-MP is regulated by drug methylation. Methylation leads to formation of non-cytotoxic metabolites and so reduces the cytotoxic effect of 6-MP. Inter-individual variations in methylation are due to the inherited activity of the enzyme thiopurine methyltransferase (TPMT). TPMT activity is governed by genotype and possibly other more complex factors. It is thus important to identify the 1 in 300 of patients who have a TPMT low homozygous genotype and who are at risk of protracted hypoplasia with minimal exposure to 6-MP. In UKALL 2011 assessment of TPMT genotype should be obtained from a local CPA accredited laboratory. **NB. It is imperative that TPMT genotype is derived from genotypic and NOT phenotypical analysis.**

Low/Low genotype patients should start mercaptopurine at 10% dose (7.5mg/m²/day or 6mg/m²/day) in Consolidation/Interim Maintenance and adjust by monthly 10% increments to maintain ANC >0.5x10⁹/L and platelets >50x10⁹/L. Continuing maintenance therapy starting dose should be the same as the dose tolerated at end of Interim Maintenance. **These patients should also receive only 10% (6mg/m²) dose of mercaptopurine during DI blocks.**

For the purpose of mercaptopurine dose adjustment, patients with **High/Low** and **High/High** genotypes will be categorised as non-variant genotype and treated the same. Additional guidance about appropriate dosing of mercaptopurine in these children may be available from the TPMT study (see Appendix 21 for details).

The oral doses of 6-MP and MTX should be adjusted to maintain ANC between 0.75x10⁹/L and 1.5x10⁹/L and platelets between 75x10⁹/L and 150x10⁹/L. This section describes the dose adjustments that should be made to maintain these levels.

Dose adjustments during treatment phases

The below table describes the dose adjustments that should be made to mercaptopurine and methotrexate during the different treatment phases for each regimen.

<u>TREATMENT PHASE</u>	<u>REGIMEN</u>	<u>DOSE ADJUSTMENT</u>
Consolidation	A	Start at 100% mercaptopurine dose (75mg/m ² /day) and do not escalate. Follow dose reduction guidelines as described below in the continuing maintenance phase. Note: mercaptopurine dose during Regimen B and C

		consolidation, delayed intensification, Protocol M and M-A commences according to blood count and is not adjusted once the treatment phase has started.
Consolidation contd.	B, C	Mercaptopurine dose during Regimen B and C consolidation is not adjusted according to blood count.
Standard Interim Maintenance	A, B	As per dose adjustment guidelines for continuing maintenance below. Do not escalate.
Protocol M / Protocol M-A	A, B, C	Mercaptopurine dose during Protocol M and Protocol M-A is not adjusted according to blood count.
Delayed Intensification	A, B, C	Mercaptopurine dose during delayed intensification is not adjusted according to blood count and these rules do not apply.
Continuing Maintenance	A, B, C	<u>DOSE ESCALATION</u> The aim is to adjust doses to maintain the ANC between 0.75 and $1.5 \times 10^9/L$ and the platelet count between 75 and $150 \times 10^9/L$. Start maintenance at 100% doses DO NOT give higher doses at the start of continuing maintenance therapy even if the patient tolerated higher doses throughout interim maintenance. During continuing maintenance if the ANC is $>1.5 \times 10^9/L$ and platelets $>150 \times 10^9/L$ for ≥ 8 weeks, the dose of mercaptopurine should be escalated by 25% (from 75mg/m²/day). Otherwise keep at 100% of dose. If the subsequent monthly ANC is $>1.5 \times 10^9/L$: 1) Keep mercaptopurine at the 125% dose and increase oral methotrexate by 25% to a dose of 25mg/m². 2) Continue to increase the mercaptopurine and oral methotrexate dose in 25% steps alternately every eight weeks as outlined above if ANC $>1.5 \times 10^9/L$ and platelets $>150 \times 10^9/L$ persists. There are no maximum doses for mercaptopurine and methotrexate. <u>DOSE REDUCTION FOR FALLING NEUTROPHIL COUNTS</u> If the neutrophil count falls to between 0.5 and $0.75 \times 10^9/L$: • HALVE the dose of mercaptopurine and oral methotrexate. If neutrophil count is $<0.5 \times 10^9/L$: • STOP mercaptopurine and methotrexate. ONLY RESTART when the count is over $0.75 \times 10^9/L$. Restart at 100% of protocol dose (not dose at which counts fell) when neutrophils $>0.75 \times 10^9/L$. NB. If counts fluctuate wildly when restarting at 100% dose, starting at 50% and titrating upwards is permissible to avoid frequent interruptions to mercaptopurine exposure. (This manoeuvre is not often necessary).

Continuing Maintenance cont.		<u>DOSE REDUCTION FOR FALLING PLATELET COUNTS</u> If the platelet count is <75 but $>50 \times 10^9/L$: • HALVE the dose of mercaptopurine and methotrexate. If platelet count is $<50 \times 10^9/L$: • STOP mercaptopurine and methotrexate. ONLY RESTART when the count is greater than $75 \times 10^9/L$. Restart at 100% of protocol dose (not dose at which counts fell) when platelet count $>75 \times 10^9/L$. NB. If counts fluctuate wildly when restarting at 100% dose, starting at 50% and titrating upwards is permissible to avoid frequent interruptions to mercaptopurine exposure. (This manoeuvre is not often necessary).
------------------------------	--	--

NOTE: Tolerance of 150% or more of the target protocol mercaptopurine dose for prolonged periods may be indicative of partial or non-compliance, and is potentially dangerous if the patient suddenly starts to comply fully. Metabolite assays in such circumstances can be helpful to exclude non-compliance. Rare individuals (1 in 300) taking mercaptopurine who are congenitally lacking intracellular TPMT will show profound myelosuppression at standard dose. These patients will be identified prospectively at the time of diagnosis (see section 7.11.2 and Appendix 21), and further advice on dosing will be given by the Chief Investigator or Co-investigators.

7.11.3 Co-trimoxazole (trimethoprim and sulphamethoxazole)

This drug is given as PCP prophylaxis orally twice a day on 2 consecutive days each week throughout all treatment phases from the start of the induction regimens. The dose is tabulated below.

Surface Area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole
0.5-0.75m ²	240mg bd	40mg bd	200mg bd
0.76-1.0 m ²	360mg bd	60mg bd	300mg bd
1.1 -1.5m ²	480mg bd	80mg bd	400mg bd
Over 1.5 m ²	480-960mg bd	80-120mg bd	400-800mg bd

*according to local policy over 1.5m²

Please ensure separation of the days on which oral methotrexate and co-trimoxazole doses are given during maintenance courses.

If a patient remains cytopenic after being off chemotherapy for three weeks or more, then stop the co-trimoxazole. Reintroduce co-trimoxazole once both mercaptopurine and methotrexate are back at standard dose. If cytopenias recur once the co-trimoxazole is reintroduced, then it should be stopped

for at least two months and an alternative form of prophylaxis used instead (see below). The alternative drug should then be continued for the duration of the antileukaemic therapy. The maintenance of adequate doses of mercaptopurine and methotrexate should take precedence over continuing co-trimoxazole. If co-trimoxazole is stopped, however, it must be remembered that the patient is at increased risk of PCP.

Alternative PCP Prophylaxis

If a patient must stop co-trimoxazole because of repeated cytopenias or other inability to tolerate it, PCP prophylaxis should continue with one of the alternative drugs. Data from HIV populations suggests that dapsone is a more effective choice than nebulised pentamidine or atovaquone. Dapsone 2mg/kg (maximum dose 100mg) once daily or 4mg/kg weekly (maximum dose 200mg), orally is recommended as the alternative agent in patients who cannot tolerate co-trimoxazole. Side effects of dapsone include fever, rash, and haemolytic anaemia. **G6PD qualitative assay should be performed before starting dapsone therapy.** For patients who cannot tolerate dapsone, nebulised pentamidine or oral atovaquone is recommended. Nebulised pentamidine 300mg per month is given by nebuliser, using 6 ml sterile water delivered at 6 L/min until the reservoir is dry, usually over 45 minutes. Atovaquone is given at a dose of 30mg/kg for patients orally once a day with food. Side effects include gastrointestinal intolerance, rash, headache, and fever. Pentamidine 4 mg/kg intravenous every 2-4 weeks could be used if none of the other alternatives are suitable.

Issue	Co-trimoxazole	Dapsone	Nebulised pentamidine	Atovaquone
Efficacy	high	moderate	moderate	moderate
Toxicity	moderate	low-moderate	high	low
Cost	low	low	high	very high
Bacterial infection protection	yes	?	no	no
Risk of extrapulmonary pneumocystosis	no	no	yes	no

7.12 Dose modifications for toxicity

The below table describes the dose alterations that should be made if a patient experiences toxicity. **This should not be used as an exclusive list and current versions of SmPCs should be referred to wherever possible.**

<u>DRUG</u>	<u>TOXICITY</u>	<u>ACTION</u>
Anthracyclines (Doxorubicin and Daunorubicin)	Hyperbilirubinemia	If total bilirubin >120 µmol/L omit dose If >90 µmol/L but ≤ 120 µmol/L give 25% of dose. If >50 µmol/L but ≤ 90 µmol/L give 50% of dose If ≤ 50 µmol/L give full dose. Check LFTs only if patient is jaundiced. Do not alter dose for abnormal transaminases.

Asparaginase	Anaphylaxis or anaphylactoid reactions:	Pegaspargase should be discontinued if the patient develops a grade 2–4 toxicity. Send blood samples to Professor Saha's laboratory (contact details at front of protocol) for asparaginase antibodies and change to crisantaspase (see Appendix 19).
	Symptomatic pancreatitis:	See Appendix 18 for diagnosis and management of pancreatitis
	Hyperglycemia	Do not modify dose. Administer insulin as required.
	Ketoacidosis:	Hold pegaspargase until blood glucose can be regulated with insulin.
	Coagulopathy	When significant symptomatic coagulopathy occurs, withhold pegaspargase until resolved. Routine clotting screens are not recommended. Coagulopathy without bleeding is not an indication to withhold pegaspargase. Management of asparaginase associated thrombosis is described in Appendix 9.
	Liver Dysfunction:	Check LFTs only if patient jaundiced. Withhold if total bilirubin >50. Do not alter dose for abnormal transaminases.
Cyclophosphamide	Prior history of gross haematuria or microscopic haematuria:	Hydrate at 125 ml/m ² /hr for 24 hours after dose and use mesna e.g. bolus doses of mesna 360mg/m ² pre, and 4, 7, 11 hours post dose or mesna 1200mg/m ² infused alongside hydration fluid or according to local established practice.
	Acute fluid retention:	Treat with frusemide and saline; do not modify dose.
Cytarabine	Hyperbilirubinaemia:	If total bilirubin >120 omit dose If >90 but ≤ 120 give 25% of dose If >50 but ≤ 90 give 50% of dose If ≤ 50 give full dose. Check LFTs only if patient jaundiced. Do not alter dose for abnormal transaminases.

Mercaptopurine	Mucositis :	For grade 2 mucositis of over 3 days duration decrease mercaptopurine dose by 30%. For grade 3-4 mucositis withhold mercaptopurine until resolved; resume at 50% of the previous dose and subsequently escalate to 75% to 100% dose at 7-14 day intervals provided grade 3-4 toxicity does not recur. Consider culturing lesions for herpes simplex if mucositis persists or recurs.
	Liver:	Check LFTs only if patient jaundiced. If bilirubin is >50 µmol/L omit mercaptopurine until it is less than 20 µmol/L, and then restart at half of the previous dose. Escalate from 50% to 75% to 100% dose at 7-14 day intervals provided hyperbilirubinaemia does not recur. Do not modify dosage for elevated aminotransferases.
Intrathecal methotrexate	Any significant neurotoxicity not due to lumbar puncture syndrome (low opening pressure, slow CSF flow, orthostatic symptoms) should be reported.	
	Systemic toxicity:	The dosage for intrathecal methotrexate will not be reduced for systemic toxicity (myelosuppression, mucositis, etc.). See Appendix 8 for management of encephalopathy related to intrathecal methotrexate.
	Viral, bacterial, or fungal meningitis:	Omit until resolved.
	Encephalopathy attributed to intrathecal methotrexate:	See Appendix 8.
Oral methotrexate	Mucositis :	For grade 2 mucositis of over 3 days duration, decrease MTX dose by 30%. For grade 3-4, mucositis, withhold MTX until resolved; resume at 50% of the previous dose and subsequently escalate to 75% to 100% dose at 7-14 day intervals provided grade 3-4 toxicity does not recur. Consider culturing lesions for herpes simplex if mucositis persists or recurs.

Oral methotrexate contd.	Liver:	Check LFTs only if patient jaundiced. If bilirubin is >50 µmol/L omit MTX until it is less than 20 µmol/L, and then restart at half of the previous dose. Escalate from 50% to 75% to 100% dose at 7-14 day intervals provided hyperbilirubinaemia does not recur. Do not modify dosage for elevated aminotransferases.
	Kidney (Grade 3-4):	Omit MTX until grade 0 toxicity (i.e. completely resolved). Resume at 100% of the previous dose and continue at 7-14 day intervals.
Intravenous methotrexate in Capizzi maintenance	Mucositis :	For grade 2 mucositis of over 3 days duration, decrease MTX dose by 30%. For grade 3-4, mucositis, withhold MTX until resolved; resume at 50% of the previously attained dose and subsequently escalate to 75% to 100% dose at 10 day intervals provided grade 3-4 toxicity does not recur. Consider culturing lesions for herpes simplex if mucositis persists or recurs.
	Liver:	If bilirubin is >50 micromoles/L omit MTX until it is less than 20 micromoles/L, and then restart at half of the previously attained dose. Escalate from 50% to 75% to 100% dose at 10-day intervals provided hyperbilirubinaemia does not recur. Do not modify dosage for elevated aminotransferases.
	Kidney (Grade 3-4):	Omit MTX until grade 0 toxicity (i.e. completely resolved). Resume at 100% of the previously attained dose and continue at 10-day intervals.
High dose methotrexate in Protocol M / M-A	Mucositis	Delay next HDMTX dose until mucositis resolved then give 100% dose. Grade 4, or recurrent Grade 3, change to standard interim maintenance. See appendix 7
	Diarrhoea	Delay next HDMTX dose until diarrhoea resolved and then give 100% dose.
	Infection	Delay until infection resolved and then give 100% dose.

High dose methotrexate in Protocol M / M-A contd.	<u>Renal function:</u>	<u>PRIOR TO STARTING FIRST PULSE OF HDMTX</u> Please note that to be eligible for entry into this phase renal function should be within normal limits corrected for age.
	Creatinine > 1.5 x baseline or GFR creatinine clearance <65L/minute/1.73m²	<u>PRIOR TO STARTING EACH SUBSEQUENT PULSE OF HDMTX</u> If renal function does not recover, omit MTX. Do not give HDMTX to a patient with this degree of renal impairment, assuming that prolonged excretion can be managed with Carboxypeptidase. Counts permitting, continue with mercaptopurine regardless of renal function throughout protocol M/M-A to 2 weeks after last course of HD MTX. Change to standard/capizzi interim maintenance if MTX excretion delayed > 168 hours, use of glucarpidase or continuing acute renal failure (<65ml/min/1.73m ² at 14 days post start of previous HDMTX dose) See appendix 7
	<u>Liver function:</u>	
	Bilirubin > upper limit of normal (ULN) for age	Delay until returns to normal.
	ALT/AST > 5xULN but <20xULN	Re-check in 36-48 hours and give next HDMTX dose if levels are decreasing and bilirubin is normal.
	ALT/AST > 20xULN	Contact Chief Investigator or Co-investigators for further advice.
	Seizures	In case of seizures which may be attributable to HDMTX contact the Chief Investigator or Co-investigators for advice.
	Neutropenia	Change to standard interim maintenance for neutrophils <0.2 x 10 ⁹ for > 7 days See appendix 7
Steroids	Hypertension:	Steroid should not be reduced. Sodium restriction and anti-hypertensives should be employed in an effort to control hypertension.
	Malignant Hypertension:	Reduce dose by 33%. Sodium restriction and antihypertensive drugs may also be utilized.

	Hyperglycemia:	Steroids should not be reduced if the patient develops clinical signs of diabetes. Rather, insulin therapy should be employed to control the blood glucose level such that symptoms and signs are minimal.
	Pancreatitis:	Do not modify dose.
	Psychosis:	Administer half dosage of steroid.
	Suspected steroid-induced myopathy:	Measure CPK with isoenzymes, consider E MG studies
	Avascular necrosis:	Contact Trial Coordinators if AVN develops before Maintenance therapy has begun. Omit further steroids if AVN develops during maintenance.
Steroids contd.	Varicella Zoster:	Steroids should be withheld during active infection except during induction (Discuss with coordinators). They should not be withheld during incubation period following exposure to varicella.
	Severe dexamethasone intolerance	Replace dexamethasone with prednisolone at a ratio of 1mg:6.67mg (e.g. 6mg/m²/day dexamethasone replaced with 40mg/m²/day prednisolone). The Trials Office should be notified of any patient switching from dexamethasone to prednisolone. Where prednisolone is used in place of dexamethasone due to severe toxicity, prednisolone will be an IMP if dexamethasone is an IMP. NB: any patient experiencing severe dexamethasone intolerance during Induction therapy must be discussed with the Chief Investigator before switching to prednisolone.
Vincristine (See also drug interactions – Appendix 6)	Seizures:	Hold 1 dose, then reinstitute.
	Severe foot drop, paresis or ileus:	Hold dose(s); institute aggressive regimen to treat constipation (except enemas if neutropenic), if present. When symptoms abate, resume at 1mg/m ² ; escalate to full dose as tolerated.
	Jaw pain:	Treat with analgesics; do not modify vincristine dose.

Hyperbilirubinemia:	Check LFTs only if patient jaundiced. Withhold if total bilirubin >50. Administer 50% of dose if total bilirubin 25 - 50. Do not alter dose for abnormal transaminases.
----------------------------	--

7.13 Supportive treatment

7.13.1 Tumour Lysis

All patients should be adequately hydrated at the start of treatment with 2-2.5L/m²/24hrs of hydration fluid appropriate to the clinical needs of the patient (e.g. glucose 2.5% with sodium chloride 0.45%). Hydration should be given parenterally for at least the first 48hrs. Potassium should not be added to the hydration fluid during induction therapy.

Allopurinol should be started 24hrs prior to induction chemotherapy and continued for at least 5 days. Rasburicase should be considered in place of allopurinol if the patient is considered at high risk of tumour lysis syndrome (e.g. white cell count >100x10⁹/L or bulky disease or elevated urate at diagnosis). See also Appendix 22 for discussion of tumour lysis syndrome.

7.13.2 Antimicrobials

All patients should receive co-trimoxazole as PCP prophylaxis which is given orally on two consecutive days each week starting from the beginning of treatment. Please give co-trimoxazole and oral methotrexate at different times of the week when both are due in the same week. Standard paediatric co-trimoxazole doses are as follows:

Surface Area	Co-trimoxazole	Trimethoprim	Sulphamethoxazole
0.5-0.75m ²	240mg bd	40mg bd	200mg bd
0.76-1.0 m ²	360mg bd	60mg bd	300mg bd
1 -1.5m ²	480mg bd	80mg bd	400mg bd
Over 1.5 m ²	480-960mg bd	80-120mg bd	400-800mg bd

*according to local policy over 1.5m²

The use of aciclovir for prophylaxis against HSV and VZV reactivation and the use of antifungal prophylaxis are not routinely recommended in paediatric practice. Adult Centres treating UKALL2011 patients may follow standard local practice. NB: azole antifungals should be avoided during treatment with vincristine.

7.13.3 Antiemetics

Prophylactic antiemetics should be administered where appropriate according to local policy and in keeping with the emetogenic potential of the treatment prescribed. Nausea and vomiting should be treated according to established local practice.

7.13.4 Mouthcare

The use of prophylactic mouthcare agents is not mandatory and is at the discretion of the treating physician.

7.13.5 G-CSF

The use of G-CSF is not routinely recommended for patients in this trial. However, in the case of life-threatening sepsis the use of G-CSF should follow local practice.

7.14 Patient Follow Up

Clinical outcomes will be measured using case reports forms (CRFs) after each course of intensive therapy and then 3-monthly whilst on therapy.

After completion of maintenance therapy, patients will be followed up using an annual questionnaire for a minimum period of five years. If a patient fails to attend clinic for any visit then the site must make every effort to obtain the requested follow-up information.

7.15 Patient Withdrawal

7.15.1 Withdrawal from UKALL 2011 trial treatment

The reason for withdrawal from UKALL 2011 protocol treatment should be recorded in the patient's medical notes and on the case report form (CRF) where it is due to either the patient's or clinician's decision. Reason for withdrawal may include, but are not limited to:

- The patient withdraws consent to further trial treatment
- The patient is not eligible for randomisation
- Unacceptable toxicity
- Disease progression whilst on therapy

UKALL 2011 will be analysed on an "intention-to-treat" basis and any patients withdrawn from trial treatment will remain within the trial unless the patient explicitly withdraws consent for data collection (see section 7.15.2)

7.15.2 Withdrawal of consent to data collection

A patient's wishes with respect to their data must be respected. If a patient explicitly states that they do not wish for any further data to be collected this must be recorded on the relevant CRF. Details should also be recorded in the patient's hospital records and no further CRFs must be completed.

7.15.3 Lost to follow-up

If a patient is lost to follow-up every effort should be made to contact the patient's GP (if consented) to obtain information on the patient's status. Similarly, if a patient's care is transferred to another clinician, the UKALL 2011 Trials Office should be informed.

7.16 Relapse

Patients who relapse in any way and at any stage while on UKALL 2011 should be taken off UKALL 2011 protocol treatment. A relapse CRF must be completed. Entry into UKALL R3 or an equivalent adult protocol should be considered.

8. ADVERSE EVENT REPORTING

The collection and reporting of Adverse Events (AEs) will be in accordance with the Medicines for Human Use Clinical Trials Regulations 2004 and its subsequent amendments. Definitions of different types of AE are listed in Appendix 2. The Investigator should assess the seriousness and causality (relatedness) of all grade 3 and 4 AEs experienced by the patient (this should be documented in the source data) with reference to the Summary of Product Characteristics.

8.1 Reporting Requirements

Reporting of adverse events will not be required for registered patients during induction and consolidation. Adverse events occurring in patients randomised to R2 treatment should be reported as detailed below.

8.1.1 Adverse Events

AEs are commonly encountered in patients receiving chemotherapy and the safety profiles of the IMPs used in this trial are well characterised, consequently data collection will focus on Common Terminology Criteria for Adverse Events (CTCAE), Grade 3 or 4 toxicities listed on the Adverse Event Form. Pre-existing conditions should only be reported if the condition worsens by at least 1 CTCAE grade. Details of all AEs experienced by the patient should be recorded in the hospital notes.

AEs (including SAEs) will not be reported for non-randomised patients.

8.1.2 Serious Adverse Events

Investigators should report AEs that meet the definition of an SAE (see Appendix 2 for definition) and are not excluded from the reporting process as described in Section 8.1.2.1 below. Such events should be reported on an **SAE Form**.

In addition, osteonecrosis should be reported on an SAE Form if it is **symptomatic** and is confirmed with supportive radiology. It should be reported as an "Other pertinent medical reason for reporting". See Appendix 17 for osteonecrosis recommendations.

8.1.2.1 Events that do not require expedited reporting

Patients receiving chemotherapy may require admission to hospital for appropriate medical intervention following development of some of the more severe known side effects of treatment. For this reason the following SAEs do not require expedited (immediate) reporting by site and are not regarded as unexpected for the purpose of this trial. These events should be reported on the **Expected Serious Adverse Event Form** upon resolution of the event.

- Any new or prolonged hospitalisation to control symptoms of **vomiting** unless the condition is life threatening or proves fatal.
- Admissions for supportive treatment during an episode of myelosuppression* unless the condition is life threatening or proves fatal.

The following are examples:

- Suspected or proven viral, bacterial, fungal or protozoal infections.
- Admissions for upper respiratory tract infection, pneumonia or gastroenteritis during therapy
- Development of cellulitis around a line site or other area

-
- Line infections (such events are considered to be expected as a result of the suppression of the marrow and lymphoid immune system and compromise of skin integrity)
 - Non-neutropenic fever
 - Mite infestations
 - Mucositis
 - Admissions for treatment of constipation
 - Admissions for treatment of allergic reactions (not requiring adrenaline or life supporting treatment)
 - Admissions for fractures
 - Admissions for post lumbar puncture pain or headache
 - Admission for simple uncomplicated seizure

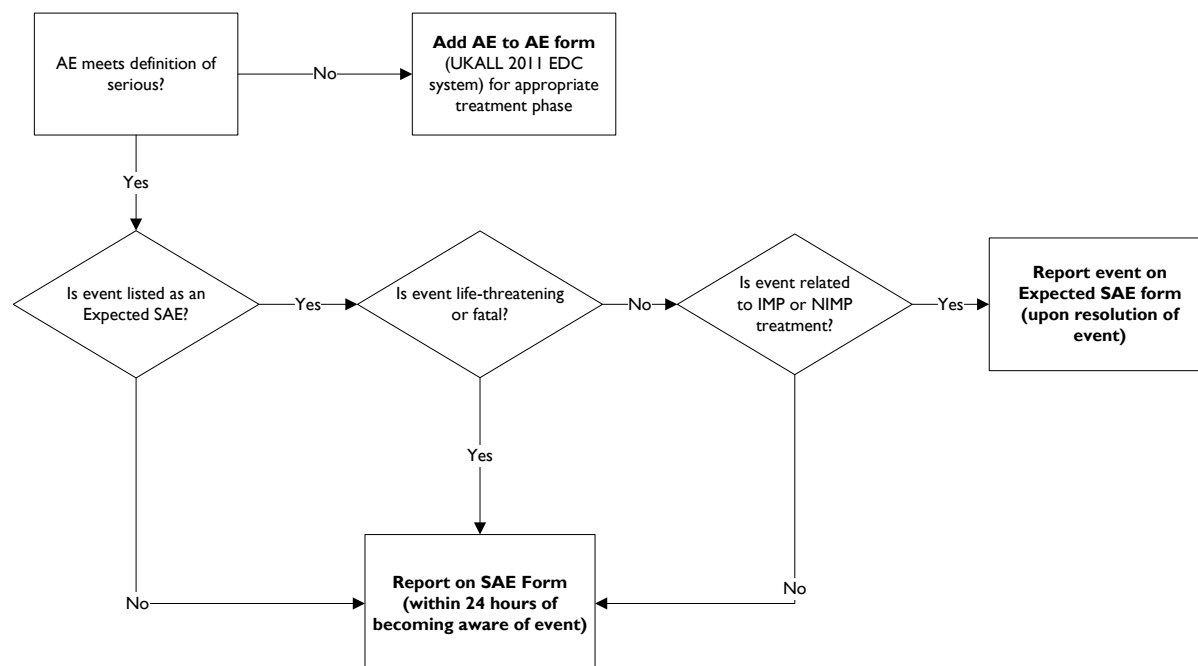
*any patient who has received chemotherapy within the protocol within the last 6 weeks

Life-threatening or fatal episodes of any of the above must be reported on an SAE Form within 24 hours of becoming aware of SAE.

An Expected SAE that falls under the category of myelosuppression should be reported as a CTCAE (v4.0) term on the Expected SAE Form

The Expected SAE Form should be used for any events defined above **occurring at any time** during a patient's trial-mandated treatment on the UKALL 2011 trial, regardless of whether the patient is receiving an IMP or a NIMP.

The following flow chart can be used to determine whether an SAE or Expected SAE Form should be completed:



8.1.2.2 Events that do not require reporting on a Serious Adverse Event Form or an Expected Serious Adverse Event Form

The following events should not be reported on an SAE Form or an Expected SAE Form:

1. Hospitalisations for:

- o Protocol defined treatment
- o Pre-planned elective procedures unless the condition worsens
- o Treatment for progression of the patient's cancer
- o Radiologic evidence of osteonecrosis in an asymptomatic patient
- o Precautionary observation
- o Supportive treatment normally administered as outpatient treatment e.g. blood transfusion but where admission to hospital has been required for logistical reasons.
- o Admission for febrile neutropenia unless life threatening (admission to ICU), leads to permanent disability or death. This data will be captured on an adverse event form

2. Progression or death as a result of the patient's cancer, as this information is captured elsewhere on the Case Report Form

8.1.2.3 Monitoring pregnancies for potential Serious Adverse Events

It is important to monitor the outcome of pregnancies of patients in order to provide SAE data on congenital anomalies or birth defects.

In the event that a trial patient becomes pregnant during the course of the trial consent must first be obtained for pregnancy monitoring using the Release of Medical Information Form. If the patient is happy to provide consent to pregnancy monitoring a Pregnancy Notification Form should be completed and returned to the Trials Office as soon as possible. A subsequent Pregnancy Notification Form should then be completed to provide the outcome. If appropriate also complete an SAE Form as detailed below.

In the event that a patient's partner becomes pregnant during the course of the trial consent must first be obtained for pregnancy monitoring. The patient should be given a Release of Medical Information Form to give to their partner. If the partner is happy to provide information on the pregnancy and outcome of their pregnancy they should sign this form. Once consent has been obtained provide details of the pregnancy on a Pregnancy Notification Form, A subsequent Pregnancy Notification Form should then be completed to provide the outcome. If appropriate also complete an SAE Form as detailed below.

SAEs related to pregnancy that are judged to be at least possibly related to the trial treatment and unexpected must still be reported in an expedited manner irrespective of how long after trial treatment administration the reaction occurred.

8.1.3 Reporting period

Details of all AEs occurring during R2 treatment phases (except those listed above) will be documented and reported from the date of commencement of protocol defined treatment until 30 days after the administration of the last treatment.

If a patient withdraws from the trial, SAEs should be reported on an SAE Form or Expected SAE Form until 30 days after receiving last trial treatment.

SAEs that are judged to be at least possibly related to the IMP(s) and unexpected must still be reported in an expedited manner irrespective of how long after IMP administration the reaction occurred.

8.2 Reporting Procedure

8.2.1 Site

8.2.1.1 Adverse Events

AEs should be reported on an AE Form (and where applicable on an SAE Form). An AE Form should be completed for each treatment phase and each cycle during maintenance.

AEs will be reviewed using the CTCAE, version 4.0 (see Appendix 3). Any AEs experienced by the patient but not included in the CTCAE should be graded by an Investigator and recorded on the AE Form using a scale of (1) mild, (2) moderate or (3) severe. For each sign/symptom, the highest grade observed since the last visit should be recorded.

8.2.1.2 Serious Adverse Events

For more detailed instructions on SAE reporting, refer to the SAE Form Completion Guidelines contained in section 5 of the Investigator Site File (ISF).

AEs defined as serious and which require reporting as an SAE (excluding events listed in Section 8.1.2.1 above) should be reported on an SAE Form. When completing the form, the Investigator will be asked to define the causality and the severity of the AE which should be documented using the CTCAE version 4.0.

On becoming aware that a patient has experienced an SAE, the Investigator (or delegate) must complete, date and sign an SAE Form. The form should be faxed together with a SAE Fax Cover Sheet to the Trials Office using one of the numbers listed below as soon as possible and no later than 24 hours after first becoming aware of the event:

To report an SAE, fax the SAE Form with an SAE Fax Cover Sheet to:

0121 414 9520 or 0121 414 3700

On receipt the Trials Office will allocate each SAE a unique reference number. This number will be transcribed onto the SAE Fax Cover Sheet which will then be faxed back to the site as proof of receipt. If confirmation of receipt is not received within 1 working day please contact the Trials Office. The SAE reference number should be quoted on all correspondence and follow-up reports regarding the SAE. The SAE Fax Cover Sheet completed by the Trials Office should be filed with the SAE Form in the ISF.

For SAE Forms completed by someone other than the Investigator, the Investigator will be required to countersign the original SAE Form to confirm agreement with the causality and severity assessments. The form should then be returned to the Trials Office in the post and a copy kept in the ISF.

Investigators should also report SAEs to their own Trust in accordance with local practice.

8.2.1.3 Provision of follow-up information

Patients should be followed up until resolution or stabilisation of the event. Follow-up information should be provided on a new SAE Form (refer to the SAE Form Completion Guidelines for further information).

8.2.2 Trials Office

On receipt of an SAE Form seriousness and causality will be determined independently by a Clinical Coordinator. An SAE judged by the Investigator or Clinical Coordinator to have a reasonable causal relationship with the trial medication will be regarded as a Serious Adverse Reaction (SAR). The Clinical Coordinator will also assess all SARs for expectedness. If the event meets the definition of a SAR that is unexpected (i.e. is not defined in the Summary of Product Characteristics) it will be classified as a Suspected Unexpected Serious Adverse Reaction (SUSAR).

8.2.3 Reporting to the Competent Authority and main Research Ethics Committee

8.2.3.1 Suspected Unexpected Serious Adverse Reactions

The Trials Office will report a minimal data set of all individual events categorised as a fatal or life threatening SUSAR to the Medicines and Healthcare products Regulatory Agency (MHRA) and main Research Ethics Committee (REC) within 7 days. Detailed follow-up information will be provided within an additional 8 days.

All other events categorised as SUSARs will be reported within 15 days.

8.2.3.2 Serious Adverse Reactions

The Trials Office will report details of all SARs (including SUSARs) to the MHRA and main REC annually from the date of the Clinical Trial Authorisation, in the form of an Annual Safety Report.

8.2.3.3 Adverse Events

Details of all AEs will be reported to the MHRA on request.

8.2.3.4 Other safety issues identified during the course of the trial

The MHRA and main REC will be notified immediately if a significant safety issue is identified during the course of the trial.

8.2.4 Investigators

Details of all SUSARs and any other safety issue which arises during the course of the trial will be reported to Principal Investigators. A copy of such correspondence should be filed in the ISF.

8.2.5 Data Monitoring Committee

The independent Data Monitoring Committee (DMC) will review all SAEs.

9. DATA HANDLING AND RECORD KEEPING

9.1 Data Collection

This trial will use an electronic data capture (EDC) system which will be used for completion of CRFs. Access to the EDC system will be granted to individuals via the Trials Office. All CRFs should be entered on to the EDC system as soon as possible but within 30 days after the phase/cycle has been completed. SAE reporting will be paper-based.

9.1.1 Registered patients data collection

Collection of minimal clinical data are required for non-randomised patients. A minimal clinical data set including: NCI risk assessment, gender, cytogenetics including cytogenetic risk group, Quality of Life (Including QuEST) and treatment regimens at induction and post remission. If available, the following data will be captured: day 8/15 marrow response, day 29 and week 14 MRD. The patients will be followed-up for relapse, death and secondary malignancy and will be followed up for five years post treatment.

9.1.2 Data collection for registered patients who decline R2

Ad hoc forms should be completed on an ongoing basis for patients declining R2. Follow up forms should be completed yearly for a minimum of 5 years starting from 1 year after treatment end. Adverse event forms and treatment forms are not required for these patients.

9.1.3 Ad hoc forms

Ad hoc forms as documented below should be completed in accordance with the protocol. It should be noted that the Serious Adverse Event Form is only applicable to patients taking part in the randomised part of the trial (R2) (see section 8.0).

Applicable to only R2 Patients

- Expected/Serious Adverse Event Form (Only applicable to patients entering R2)

Applicable to all patients

- Relapse Form
- Death Report Form
- Secondary Tumour Form
- Transplant Form

-
- Pregnancy Notification Form
 - Ineligible/Declined Form

The CRF must be completed (and for the paper SAE Form signed/dated and faxed to the Trials Office) by the Investigator or an authorised member of the site research team (as delegated on the Site Signature and Delegation Log) within the timeframe listed above.

Entries on paper CRFs should be made in ballpoint pen, in blue or black ink, and must be legible. Any errors should be crossed out with a single stroke, the correction inserted and the change initialled and dated. If it is not obvious why a change has been made, an explanation should be written next to the change.

Data reported on each form should be consistent with the source data or the discrepancies should be explained. If information is not known, this must be clearly indicated on the form. All missing and ambiguous data will be queried. All sections are to be completed before being submitted.

In all cases it remains the responsibility of the Investigator to ensure that the CRF has been completed correctly and that the data are accurate.

Where any paper CRFs are used the completed originals should be sent to the Trials Office and a copy filed in the Investigator Site File.

Trial forms may be amended by the Trials Office, as appropriate, throughout the duration of the trial. Whilst this will not constitute a protocol amendment, sites will be notified of new versions of the form when they are available in the EDC system, and in the case of the SAE Form, new versions of the form must be implemented by participating sites immediately on receipt.

9.2 Archiving

It is the responsibility of the Principal Investigator to ensure all essential trial documentation and source records (e.g. signed Informed Consent Forms, Investigator Site Files, Pharmacy Files, patients' hospital notes, copies of CRFs etc) at their site are securely retained for at least 5 years after the end of the trial. Do not destroy any documents without prior approval from the CRCTU Document Storage Manager.

10. QUALITY MANAGEMENT

10.1 Site Set-up and Initiation

All sites will be required to sign a Clinical Study Site Agreement prior to participation. In addition all participating Investigators will be asked to sign the necessary agreements and supply a current CV to the Trials Office. All members of the site research team will also be required to sign the Site Signature and Delegation Log, which should be returned to the Trials Office. Prior to commencing recruitment all sites will undergo a process of initiation. Key members of the site research team will be required to attend either a meeting or a teleconference covering aspects of the trial design, protocol procedures, Adverse Event reporting, collection and reporting of data and record keeping. Sites will be provided with an Investigator Site File and a Pharmacy File containing essential documentation, instructions, and other documentation required for the conduct of the trial. The Trials Office must be informed immediately of any change in the site research team.

10.2 On-site Monitoring

Monitoring will be carried out as required following a risk assessment and as documented in the UKALL 2011 Quality Management Plan. Additional on-site monitoring visits may be triggered for example by poor CRF return, poor data quality, low SAE reporting rates, excessive number of patient withdrawals or deviations. If a monitoring visit is required the Trials Office will contact the site to

arrange a date for the proposed visit and will provide the site with written confirmation. Investigators will allow the UKALL 2011 trial staff access to source documents as requested.

10.3 Central Monitoring

Where a patient has given explicit consent sites are requested to send in copies of signed Informed Consent Forms for in-house review.

Trials staff will be in regular contact with the site research team to check on progress and address any queries that they may have. Trials staff will check incoming Case Report Forms for compliance with the protocol, data consistency, missing data and timing. Sites will be sent Data Clarification Forms requesting missing data or clarification of inconsistencies or discrepancies.

Sites may be suspended from further recruitment in the event of serious and persistent non-compliance with the protocol and/or GCP, and/or poor recruitment. Any major problems identified during monitoring may be reported to the Trial Management Group, Trial Steering Committee and the relevant regulatory bodies. This includes reporting serious breaches of GCP and/or the trial protocol to the main Research Ethics Committee (REC) and the Medicines for Healthcare products Regulatory Agency (MHRA).

10.4 Audit and Inspection

The Investigator will permit trial-related monitoring, audits, ethical review, and regulatory inspection(s) at their site, providing direct access to source data/documents.

Sites are also requested to notify the Trials Office of any MHRA inspections.

10.5 Notification of Serious Breaches

In accordance with Regulation 29A of the Medicines for Human Use (Clinical Trials) Regulations 2004 and its amendments the Sponsor of the trial is responsible for notifying the licensing authority in writing of any serious breach of:

- The conditions and principles of GCP in connection with that trial or;
- The protocol relating to that trial, within 7 days of becoming aware of that breach

For the purposes of this regulation, a “serious breach” is a breach which is likely to affect to a significant degree:

- The safety or physical or mental integrity of the subjects of the trial; or
- The scientific value of the trial

Sites are therefore requested to notify the Trials Office of a suspected trial-related serious breach of GCP and/or the trial protocol. Where the Trials Office is investigating whether or not a serious breach has occurred sites are also requested to cooperate with the Trials Office in providing sufficient information to report the breach to the MHRA where required and in undertaking any corrective and/or preventive action.

11. END OF TRIAL DEFINITION

For the purposes of main REC and MHRA approval, the trial end date is deemed to be 12 months after last data capture following a minimum of 5 years follow-up after administration of last trial treatment.

The Trials Office will notify the MHRA and main REC that the trial has ended at the appropriate time and will provide them with a summary of the clinical trial report within 12 months of the end of trial.

12. STATISTICAL CONSIDERATIONS

12.1 Randomisation

This is a prospective randomised controlled clinical trial (RCT). Randomisation will be achieved by computer programme.

Randomisation will be performed by computer programme, with balance over key variables (disease type, sex, age, WCC) achieved by the method of minimisation. The second randomisation will also be balanced by randomised steroid and MRD risk.

12.2 Arrangements for protecting against bias

All analyses will be based on intention-to-treat. Blinding is not possible, but the clinical outcomes are objectively defined, with relapse only defined as an event if it requires intensive relapse therapy. Where necessary, outcomes will be reviewed by an assessor blind to the treatment allocation. Records of the reasons for non-return of HRQoL questionnaires will be maintained and used, together with sensitivity testing, to examine for possible bias due to missing data.

12.3 Definition of Outcome Measures

12.3.1 Primary outcome measures

The primary outcome measures are:

1. Dexamethasone Randomisation (R1)

Induction steroid-induced morbidity and mortality defined as all serious, grade 3 or 4 adverse events related to induction and categorised as steroid related or steroid contributory and all induction treatment related deaths during induction.

2. Methotrexate Randomisation (R2)

Central nervous system (CNS) relapse, defined as any relapse with CNS involvement, including combined.

3. Pulses randomisation (R2)

Bone marrow relapse, defined as any relapse with bone marrow involvement, including combined, Quality of Life measured by PedsQL.

4. Overall trial population

Event free and overall survival, relapse rate, treatment related mortality and morbidity compared with UKALL 2003.

Any event, defined as relapse, secondary tumour or death from any cause, is also a primary outcome measure for each randomised comparison, and for the trial overall.

Clinical outcomes will be measured using internationally accepted definitions of treatment-related mortality (TRM) and NCI CTCAE toxicity grades (see Appendix 3 for CTCAE version 4.0). [Relapse will be diagnosed on the basis of morphology \(>= M2 bone marrow or blasts at extramedullary site\) confirmed by either flow cytometry, immunohistochemistry or cytogenetics.](#) For the dexamethasone randomisation, steroid induced morbidity in induction will be assessed as any induction treatment related death and all serious adverse events (SAEs), within 8 weeks of the start of induction, which are classified as steroid related or contributory. I.e. events reported on the Steroid AE form or classed as possibly, probably or definitely related to dexamethasone given in induction on an SAE form. Steroid related AE refers to any events known to be directly caused by exposure to high dose glucocorticoids namely diabetes, psychosis, hypertension, obesity and pathological osteopenia or fracture. Steroid contributory refers to those in which exposure to steroid is a significant contributory factor namely severe bacterial, fungal or viral infection, pancreatitis, thrombosis and encephalopathy. Quality of Life (QoL) will be assessed using questionnaires, originally designed by Varni, to score generic and disease specific measures in a paediatric population validated in UKALL2003 (see Appendix 13).

12.3.2 Secondary outcome measures

All the following are secondary outcomes except as specified above.

- Total mortality.
- Isolated BM relapse.

-
- Any BM relapse.
 - Non-BM relapse.
 - Isolated CNS relapse.
 - Any CNS relapse.
 - Non-CNS relapse.
 - Any relapse.
 - Secondary tumour.
 - Induction death.
 - Rate of remission
 - Death in remission.
 - Quality of life.
 - Day 29 MRD level.
 - Toxicity.

12.4 Analysis of Outcome Measures

The main hypotheses to be tested are:

- (a) Short (higher dose) dexamethasone for 14 days reduces steroid related toxicity compared with standard dose dexamethasone for 28 days.
- (b) High dose methotrexate compared with protracted intrathecal therapy reduces the CNS relapse rate in ALL, as measured by the time from start of induction therapy to any relapse with CNS involvement.
- (c) Treatment without vincristine plus dexamethasone pulses in maintenance improves quality of life but does not materially increase BM relapses (ALL) or local relapse (LBL).
- (d) Non-randomised changes to treatment backbone compared with UKALL 2003 have not resulted in inferior outcomes.

Patients in whom informed consent has been withdrawn, or who are classified as misdiagnosis, with a clear definition of the alternative diagnosis, or who are found during induction to have mature B-ALL or Ph positive ALL, will be excluded from all analyses. Otherwise, all main analyses will be intention-to-treat. Patients who go off protocol because they fail induction will be censored at that time in analyses of toxicity, but not of other outcomes. Main treatment comparisons will be unstratified.

Analyses of relapse, secondary tumours, death in remission, any event and overall mortality will use Kaplan-Meier curves, censoring at competing events and compared using logrank tests. 95% confidence intervals of the hazard ratios will be calculated. Steroid toxicity, induction mortality rates and day 29 MRD risk level will be compared using chi-square tests. Differences in quality of life mean values at 18 months will be tested by a two-sample t-test. Comparisons will be considered as significant using a 2-sided p-value of 0.05.

12.5 Planned Sub Group Analyses

Treatment effects will be investigated in the following subgroups: ALL, LBL, males, females; age <10 years, 10-15 years, ≥16 years old; NCI risk groups; day 29 MRD risk groups, lineage (B, T); other treatment allocations. Results will be presented as forest plots of the treatment effects with heterogeneity tests between subgroups.

12.6 Planned Interim Analysis

Interim analyses of the primary outcomes for main treatment comparisons will be presented annually to the Data Monitoring Committee (DMC). For the primary outcomes, the first analysis for the

dexamethasone comparison will be after one year of recruitment, and after 4 years for the other comparisons. The first time to event analysis will be at 4 years. Induction and remission deaths will be presented in detail from the end of year one.

In addition to overall comparisons, the dexamethasone comparison will be analysed within age subgroups (<10 years, ≥10 years).

12.7 Stopping Guidelines

In the light of the interim analyses, the DMC will advise the Steering Committee if, in their view, the randomised comparisons in the trial have provided proof beyond reasonable doubt ($2P < 0.001$ for a primary outcome difference) that for all or for some types of patient one treatment is clearly indicated or clearly contradicted. If one of the randomisations is suspended or closed, the other randomisations will continue.

Safety monitoring will be guided by the numbers of deaths in remission. If there is a difference of more than 3% for the dexamethasone or methotrexate randomisations, or an increase of more than 3% in the no pulses arm, the DMC will consider advising the Steering Committee to close a randomisation, after detailed examination of the causes of death. They may, however, recommend alternative protocol amendments.

12.8 Planned Final Analyses

The main analyses will be 2 years after closure of recruitment. Funding and regulations permitting, long term follow-up will continue and effects at 10 and 15 years analysed.

Compliance will be monitored and reported, including reasons wherever possible.

12.9 Power Calculations

The **dexamethasone randomisation** was open to all 2640 entrants. In UKALL 2003, the rate of steroid-related toxicity was 8%. Allowing for up to 10% drop out or refusal of the randomisation ($n=2376$) there will be 89% power to detect a 3.2% reduction to 4.8% (risk ratio = 0.6). The dexamethasone randomisation was closed (31-March-2017) on the advice of the DMC.

The **methotrexate randomisation** will be open to all 2376 except those with persistent high level MRD at 14 weeks (ALL patients) i.e. patients who are considered MRD High Risk (from UKALL 2003 data is expected to be 2.5% of patients) as well as the 25% of patients with LBL who fail to have an adequate response at the end of induction. Allowing a further 20% loss of patients ineligible for randomisation for clinical reasons or patient/parent choice gives around 1816 ALL randomised. At present in UKALL 2003 the CNS relapse rate is 4%, the majority in the second and third years. There will be 90% power to detect a decrease in this rate from 4% to 1.5% (relative hazard 0.37) by log-rank testing, and 86% power to detect a decrease from 3% to 1.0% (relative hazard 0.33) if the control rate proves to be lower.

The **pulses randomisation** will include the 1816 randomised above. However, 59 of these will be LBL patients for whom the BM relapse outcome measure is not applicable. In a new meta-analysis of vincristine/steroid pulses, the effect of pulses in older trials was an odds reduction of 40% in bone marrow relapses, whereas there was no evidence of an effect in the more recent trials, with an odds ratio of 0.97 (95% CI = 0.81-1.16). The more recent trial protocols were more intensive, with a 5-year event free survival of around 80% with or without pulses, compared to 70% (with pulses) and 62% (without) in the older trials. The BM relapse rate in the more recent trials was similar to the 15% seen in ALL97, and at present it is somewhat lower in UKALL 2003. Assuming a BM relapse rate of around 10% to 15%, 1757 patients would give over 85% power to rule out a 5% increase.

The **HRQoL study** piloted in UKALL 2003 shows a drop of more than one standard deviation in the mean values for the three main measures (physical, emotional and social functioning) at the end of intensification, increasing back by 50-80% by the end of treatment. Should this improvement occur earlier in the no-pulses arm, measurement at around 18 months will show a difference of 0.25-0.4

standard deviations (sd). To detect 0.25 SD would require 340 patients per treatment group to achieve 90% power from a crude comparison between randomised treatments at this time point, and 250 per group for 80% power.

13. TRIAL ORGANISATIONAL STRUCTURE

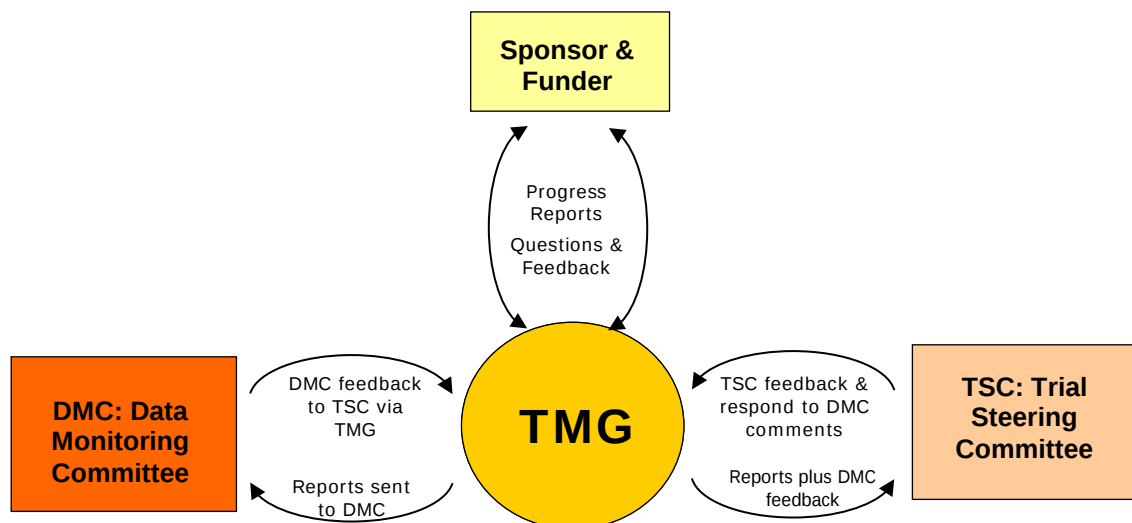
13.1 Sponsor

This study is sponsored by The University of Birmingham.

13.2 Coordinating Centre

The trial is being conducted under the auspices of the Cancer Research UK Clinical Trials Unit (CRCTU), University of Birmingham according to their local procedures.

13.3 Relationship of trial committees



13.4 Trial Management Group

The Trial Management group (TMG) is composed of the Chief Investigator, Co-investigators and the trial team at the CRCTU. The TMG is responsible for the day-to-day running and management of the trial and will meet by teleconference or in person as required.

13.5 Trial Steering Committee

The Trial Steering Committee (TSC) will provide overall supervision for the trial and provide advice through its independent chair. Membership includes independent clinicians, the trial statistician plus the Chief Investigator and members of the TMG as appropriate. The ultimate decision for the continuation of the trial lies with the TSC. The TSC will meet at least once a year or more often if required, either in person or by teleconference.

13.6 Data Monitoring Committee

Data analyses will be supplied in confidence to an independent Data Monitoring Committee (DMC), which will be asked to give advice on whether the accumulated data from the trial, together with the results from other relevant research, justifies the continuing recruitment of further patients. The DMC will operate in accordance with a trial specific charter based upon the template created by the Damocles Group. During the recruitment phase of the trial the DMC is scheduled to meet within the six months of the first site opening to recruitment and annually thereafter. Additional meetings will be held if required. Additional meetings may be called if recruitment is much faster than anticipated and the DMC may, at their discretion, request to meet more frequently or continue to meet following completion of recruitment. An emergency meeting may also be convened if a safety issue is identified. The DMC will report directly to the Trial Management Group (TMG) who will convey the findings of the DMC to Trial Steering Committee, MHRA, funders, and sponsors where applicable. The DMC may consider recommending the discontinuation of the trial if the recruitment rate or data quality are unacceptable or if any issues are identified which may compromise patient safety. The trial would also stop early if the interim analyses showed differences between treatments that were deemed to be convincing to the clinical community. See section 12.7 for stopping rules.

13.7 Finance

This is a clinician-initiated and clinician-led trial funded by Leukaemia and Lymphoma Research. No individual per patient payment will be made to NHS Trusts, Investigators or patients.

This study has been adopted into the NIHR CRN Portfolio.

14. ETHICAL CONSIDERATIONS

The trial will be performed in accordance with the recommendations guiding physicians in biomedical research involving human subjects, adopted by the 18th World Medical Association General Assembly, Helsinki, Finland, June 1964, amended at the 48th World Medical Association General Assembly, Somerset West, Republic of South Africa, October 1996 (website: <http://www.wma.net/en/30publications/10policies/b3/index.html>). See also Appendix 1 for Declaration of Helsinki.

The trial will be conducted in accordance with the Research Governance Framework for Health and Social Care, the applicable UK Statutory Instruments, (which include the Medicines for Human Use Clinical Trials 2004 and subsequent amendments and the Data Protection Act 1998 and Human Tissue Act 2008" if appropriate and the International Conference on Harmonisation Guidelines for Good Clinical Practice (ICH GCP). This trial will be carried out under a Clinical Trial Authorisation in accordance with the Medicines for Human Use Clinical Trials regulations. The protocol will be submitted to and approved by the main Research Ethics Committee (REC) prior to circulation.

Before any patients are enrolled into the trial, the Principal Investigator at each site is required to obtain local R&D approval. Sites will not be permitted to enrol patients until written confirmation of R&D approval is received by the Trials Office.

It is the responsibility of the Principal Investigator to ensure that all subsequent amendments gain the necessary local approval. This does not affect the individual clinicians' responsibility to take immediate action if thought necessary to protect the health and interest of individual patients.

15. CONFIDENTIALITY AND DATA PROTECTION

Personal data recorded on all documents will be regarded as strictly confidential and will be handled and stored in accordance with the Data Protection Act 1998. With the patient's consent, their full name, date of birth, National Health Service (NHS) number, or in Scotland the Community Health Index (CHI), hospital number and general practitioner details will be collected at trial entry to allow tracing through the Cancer Registries and the NHS Information Centre for Health and Social Care (service formally provided by the Office of National Statistics) and to assist with long-term follow-up via other health care professionals (e.g. patient's GP). Patients will be identified using only their unique trial number, hospital number, initials and date of birth on the Case Report Form and correspondence between the Trials Office and the participating site. However patients are asked to give permission for the Trials Office to be sent a copy of their signed Informed Consent Form which will not be anonymised. This will be used to perform in-house monitoring of the consent process and may also be forwarded to other health care professionals involved in the treatment of the patient (e.g. patient's GP).

The Investigator must maintain documents not for submission to the Trials Office (e.g. Patient Identification Logs) in strict confidence. In the case of specific issues and/or queries from the regulatory authorities, it will be necessary to have access to the complete trial records, provided that patient confidentiality is protected.

The Trials Office will maintain the confidentiality of all patients' data and will not disclose information by which patients may be identified to any third party other than those directly involved in the treatment of the patient and organisations for which the patient has given explicit consent for data transfer. Representatives of the UKALL 2011 trial team may be required to have access to patient's notes for quality assurance purposes but patients should be reassured that their confidentiality will be respected at all times.

16. INSURANCE AND INDEMNITY

University of Birmingham employees are indemnified by the University insurers for negligent harm caused by the design or coordination of the clinical trials they undertake whilst in the University's employment.

In terms of liability at a site, NHS Trust and non-Trust hospitals have a duty to care for patients treated, whether or not the patient is taking part in a clinical trial. Compensation is therefore available via NHS indemnity in the event of clinical negligence having been proven.

The University of Birmingham cannot offer indemnity for non-negligent harm. The University of Birmingham is independent of any pharmaceutical company, and as such it is not covered by the Association of the British Pharmaceutical Industry (ABPI) guidelines for patient compensation.

17. PUBLICATION POLICY

Results of this trial will be submitted for publication in a peer reviewed journal. The manuscript will be prepared by the Trial Management Group (TMG) and authorship will be determined by mutual agreement.

Any secondary publications and presentations prepared by Investigators must be reviewed by the TMG. Manuscripts must be submitted to the TMG in a timely fashion and in advance of being submitted for publication, to allow time for review and resolution of any outstanding issues. Authors must acknowledge that the trial was performed with the support of The University of Birmingham. Intellectual property rights will be addressed in the Clinical Study Site Agreement between Sponsor and site.

18. REFERENCE LIST

1. Medical Research Council UKALL Trials 1972–84. Improvement in treatment for children with acute lymphoblastic leukaemia. Report to the council by the Working Party on Leukaemia in Childhood. *Lancet* 1986; i: 408–411
2. Eden OB, Lilleyman JS, Richards S, Shaw MP, Peto J. Results of the MRC Leukaemia Trial UKALL VIII. *Br J Haematol* 1991; 78:187–196
3. Chessells JM, Bailey CC et al . Intensification of treatment and survival in all children with lymphoblastic leukaemia; results of UK Medical Research Council trial UKALL X. *Lancet*. 1995;345:143-148.
4. Hill F, Richards S et al Successful treatment without cranial radiotherapy of children receiving intensified chemotherapy for acute lymphoblastic leukaemia: results of risk stratified randomized central nervous system treatment trial MRC UKALL XI: *B J Haem* 124 33-46 2004
5. Hann I, Vora A, et al. Benefit of intensified treatment for all children with acute lymphoblastic leukaemia: results from MRC UKALL XI and MRC ALL97 randomised trials. UK Medical Research Council's Working Party on Childhood Leukaemia. *Leukemia*. 2000;14:356-363
6. Mitchell C, Payne J et al. The impact of risk stratification by early bone marrow response in childhood acute lymphoblastic leukaemia; results from the United Kingdom Medical Research Council trial ALL 97 and ALL 97/99 *Br J Haem* 146 424-436 2009
7. Nachman J, Sather H et al Augmented post induction therapy for children with high risk ALL and slow response to initial therapy. *NEJM*, 338 1663, 1998
8. Vora AJ, Mitchell C et al UKALL 2003, A Randomised Trial Investigating Treatment Reduction for Children and Young Adults with Minimal Residual Disease Defined Low Risk Acute Lymphoblastic Leukaemia *Blood* 2010 November 116; 496
9. Reiter A, Schrappe M et al Intensive ALL-type therapy without local radiotherapy provides a 90% event-free survival for children with T-cell lymphoblastic lymphoma: a BFM Group report. *Blood* 95 416-421 2000
10. Burkhardt B, Woessman W et al Impact of cranial radiotherapy on central nervous system prophylaxis in children and adolescents with central nervous system-negative stage III or IV lymphoblastic lymphoma. *JCO* 24 491-9 2006.
11. Siebel N, Steinherz P et al Early postinduction intensification therapy improves survival for children and adolescents with high-risk acute lymphoblastic leukemia: a report from the Children's Oncology Group. *Blood*. 2008;111:2548-2555
12. Matloub Y , Angiolillo A, et al Double Delayed Intensification (DDI) Equivalent to Single DI (SDI) in Children with National Cancer Institute (NCI) Standard-Risk Acute Lymphoblastic Leukemia SR-ALL) Treated on Children's Cancer Group (CCG) Clinical Trial 1991 (CCG-1991) *Blood* (ASH Annual Meeting Abstracts), Nov 2006; 108: 146
13. Pui C, Campana D et al. Treating ALL without Cranial Irradiation. *NEJM* 360, 2730-2741 2009
14. Schrappe M , Zimmerman M et al. Dexamethasone in induction can eliminate one third of all relapses in childhood Acute Lymphoblastic Leukaemia: results of an international randomised trial in 3655 patients (Trial AIEOP BFM ALL 2000) *Blood* (ASH Annual Meeting Abstracts), Nov 2008; 112:
15. Balduzzi A, Valsecchi M. Relapse in Childhood "Very High Risk" Acute Lymphoblastic Leukemia Treated with BFM Chemotherapy Has a Dismal Outcome Even When Treated with Transplantation. *Blood* (ASH Annual Meeting Abstracts), Nov 2008; 112: 91
16. Gaynon P, Harris RE et al Bone marrow transplantation versus prolonged intensive chemotherapy for children with acute lymphoblastic leukaemia and an initial bone marrow relapse within 12 months of the completion of primary therapy: Children's Oncology Group study CCG-1941 *JCO*; 24(19):3150-6, 2006.
17. Sallan SE Myths and Lessons from the adult paediatric interface. *Hematology* (ASH education program) 2006
18. Schrauder A, Stanulla M et al. Prospective evaluation of MRD-Kinetics in 274 children with high risk ALL treated in trial ALL BFM 2000: insights into development of resistance and impact on further refinement of treatment stratification *Blood* 109 585
19. Mc Neer JL, Nachman J The optimal use of steroids in paediatric acute lymphoblastic leukaemia: no easy answers. *B J Haem* 149 638 -52 2010

20. Mitchell C, Richards S et al. Benefit of dexamethasone compared with prednisolone for childhood acute lymphoblastic leukaemia: results of the United Kingdom Medical Research Council trial ALL 97 and ALL 97/99 Br J Haem 129 734 -745 2005
21. Bostrom BC, Gaynon P, *et al.* Dexamethasone (DEX) decreases central nervous system (CNS) relapse and improves event-free survival (EFS) in lower risk acute lymphoblastic leukemia (ALL). Proc Am Soc Clin Oncol. 1998;17:527a.
22. Moricke A, Zimmerman M et al No influence on incidence of osteonecrosis when dexamethasone replaces prednisolone in induction treatment for childhood ALL. Results of BFM 2000 Blood (ASH Annual Meeting Abstracts), Nov 2008; 112: 899
23. Mattano L, Nachman J et al Increased incidence of osteonecrosis with a dexamethasone induction for high risk ALL; a report from the Children's Oncology Group Blood (ASH Annual Meeting Abstracts), Nov 2008; 112: 898
24. Lawson SE, Harrison G UK experience of treating relapsed childhood ALL, MRC UKALL R1 trial. B J Haem 108 531 2000
25. Schrappe M, Reiter A et al Long-term results of four consecutive trials in childhood ALL performed by the ALL-BFM study group from 1981 to 1995. Berlin-Frankfurt-Münster BFM Leukemia 14 2205 2000.
26. Schrappe M, Reiter A et al Improved outcome in childhood acute lymphoblastic leukemia despite reduced use of anthracyclines and cranial radiotherapy: results of trial ALL-BFM 90. German-Austrian-Swiss ALL-BFM Study Group. BFM 95, Blood 95, 3310, 2000
27. Clarke M Gaynon P et al. Childhood ALL Collaborative Group. CNS-directed therapy for childhood acute lymphoblastic leukaemia: overview of 43 randomised trials involving 13,162 children. Journal of Clinical Oncology. 21 1798-1809.
28. Pui CH, Campana D et al . Treating childhood acute lymphoblastic leukaemia without cranial irradiation. NEJM 360 2730-41 2009
29. Krishnan R, Wade S et al. Temporal changes in incidence and pattern of central nervous system relapses in children with acute lymphoblastic leukaemia: The United Kingdom experience 1985-2001. Leukemia in Press
30. Masurekar A, Parker C et al Mitoxantrone Improves the Outcome of Children with Central Nervous System (CNS) Involvement at First Relapse of Acute Lymphoblastic Leukemia (ALL)-Results of the International ALLR3 Study Blood 116 3303 2010.
31. Abromowitch M, Termuhlen A, Chang M, et al.: High-dose methotrexate and early intensification of therapy do not improve 3 year EFS in children and adolescents with disseminated lymphoblastic lymphoma: results of the randomized arms of COG A5971. [Abstract] Blood 112 (11): A-3610, 2008.
32. Matloub Y, Bostrom B et al. Escalating dose intravenous Methotrexate without leucovorin rescue during interim maintenance is superior to oral methotrexate for children with standard risk ALL. Children's Oncology Group study 1991. Blood (ASH Annual Meeting Abstracts), Nov 2008; 112: 9
33. Conter V, Valsecchi M et al, Pulses of vincristine and dexamethasone in addition to intensive chemotherapy for children with intermediate risk ALL; a multicentre randomised trial. Lancet 2007, 369 123-129.
34. Eden O, Pieters R et al Systematic review of the addition of vincristine plus steroid pulses in maintenance treatment for childhood acute lymphoblastic leukaemia - an individual patient data meta-analysis involving 5,659 children. BJ Haem 149 722-33 2010
35. De Moerloose B, Suciu S Improved outcome with pulses of vincristine and corticosteroids in continuation therapy of children with average risk acute lymphoblastic leukemia (ALL) and lymphoblastic non-Hodgkin lymphoma (NHL): report of the EORTC randomized phase 3 trial 58951. Blood 116 36-44 2010
36. van Dongen JJ, Seriu T, Panzer-Grumayer ER, et al. Prognostic value of minimal residual disease in acute lymphoblastic leukaemia in childhood. Lancet. 1998;352:1731-1738.
37. Conter V, Bartram C et al Molecular response to treatment redefines all prognostic factors in children and adolescents with B-cell precursor acute lymphoblastic leukemia: results in 3184 patients of the AIEOP-BFM ALL 2000 study Blood 2010 115:3206-3214;
38. Matloub Y, Gaynon P *et al* Bone Marrow (BM) Minimal Residual Disease (MRD) at End of Induction and Interim Maintenance Is Highly Predictive of Outcome in Children with Standard Risk (SR) Acute Lymphoblastic Leukemia (ALL) Treated on the Children's Oncology Group Study 1991. Blood (ASH Annual Meeting Abstracts), Nov 2008; 112: 701.

-
39. Borowitz M, Devidas M et al Clinical significance of minimal residual disease in childhood acute lymphoblastic leukaemia and its relationship to other prognostic factors a Children's Oncology Group study. *Blood* 111, 5477-5485 2008
 40. Rattei R, Basso G et al Monitoring treatment response of childhood precursor B-cell acute lymphoblastic leukemia in the AIEOP-BFM-ALL 2000 protocol with multiparameter flow cytometry: predictive impact of early blast reduction on the remission status after induction. *Leukemia* 23 528 2000
 41. Flohr T, Scrauder A et al. Minimal residual disease-directed risk stratification using real-time quantitative PCR analysis of immunoglobulin and T-cell receptor gene rearrangements in the international multicenter trial AIEOP-BFM ALL 2000 for childhood acute lymphoblastic leukemia. *Leukemia* 22 771 2008
 42. Zhou J, Goldwasser M Quantitative analysis of MRD predicts outcome in BCP ALL in DFCI 95-01. *Blood* 110, 1607-1611 , 2007
 43. Moghrabi A, Levy D et al Results of DFCI ALL consortium protocol 95-01 for children with acute lymphoblastic leukaemia. *Blood* 109 896-904 2007
 44. Childhood ALL collaborative group. Duration and intensity of maintenance chemotherapy in ALL: overview of 42 trials involving 12000 randomised children. *Lancet* 347 1783 1996
 45. Moorman AV, Ensor H et al Prognostic effect of chromosomal abnormalities in childhood B-cell precursor acute lymphoblastic leukaemia: results from the UK Medical Research Council ALL97/99 randomised trial. *Lancet Oncol* 11 429 -38 2010
 46. Winick n, Salzer W, Devidas M et al Dexamethasone v Prednisolone during induction for high risk ALL 0 A report from COG 0232. *JCO Vol* 29 May 20 supplement 2011 abstract 9504
 47. Strother DR, Glynn-Barnhart A, Kovnar E, Gregory RE, Murphy SB. Variability in the disposition of intraventricular methotrexate: a proposal for rational dosing. *J Clin Oncol.* 1989;7(11):1741-1747.

APPENDIX 1 - WMA DECLARATION OF HELSINKI**WORLD MEDICAL ASSOCIATION DECLARATION OF HELSINKI****Recommendations guiding physicians
in biomedical research involving human subjects**

Adopted by the 18th World Medical Assembly

Helsinki, Finland, June 1964

and amended by the

29th World Medical Assembly, Tokyo, Japan, October 1975

35th World Medical Assembly, Venice, Italy, October 1983

41st World Medical Assembly, Hong Kong, September 1989

and the

48th General Assembly, Somerset West, Republic of South Africa, October 1996

INTRODUCTION

It is the mission of the physician to safeguard the health of the people. His or her knowledge and conscience are dedicated to the fulfilment of this mission.

The Declaration of Geneva of the World Medical Association binds the physician with the words, "The Health of my patient will be my first consideration," and the International Code of Medical Ethics declares that, "A physician shall act only in the patient's interest when providing medical care which might have the effect of weakening the physical and mental condition of the patient."

The purpose of biomedical research involving human subjects must be to improve diagnostic, therapeutic and prophylactic procedures and the understanding of the aetiology and pathogenesis of disease.

In current medical practice most diagnostic, therapeutic or prophylactic procedures involve hazards. This applies especially to biomedical research.

Medical progress is based on research which ultimately must rest in part on experimentation involving human subjects.

In the field of biomedical research a fundamental distinction must be recognized between medical research in which the aim is essentially diagnostic or therapeutic for a patient, and medical research, the essential object of which is purely scientific and without implying direct diagnostic or therapeutic value to the person subjected to the research.

Special caution must be exercised in the conduct of research which may affect the environment, and the welfare of animals used for research must be respected.

Because it is essential that the results of laboratory experiments be applied to human beings to further scientific knowledge and to help suffering humanity, the World Medical Association has prepared the following recommendations as a guide to every physician in biomedical research involving human subjects. They should be kept under review in the future. It must be stressed that the standards as drafted are only a guide to physicians all over the world. Physicians are not relieved from criminal, civil and ethical responsibilities under the laws of their own countries.

I. BASIC PRINCIPLES

1. Biomedical research involving human subjects must conform to generally accepted scientific principles and should be based on adequately performed laboratory and animal experimentation and on a thorough knowledge of the scientific literature.
2. The design and performance of each experimental procedure involving human subjects should be clearly formulated in an experimental protocol which should be transmitted for

consideration, comment and guidance to a specially appointed committee independent of the investigator and the sponsor provided that this independent committee is in conformity with the laws and regulations of the country in which the research experiment is performed.

3. Biomedical research involving human subjects should be conducted only by scientifically qualified persons and under the supervision of a clinically competent medical person. The responsibility for the human subject must always rest with a medically qualified person and never rest on the subject of the research, even though the subject has given his or her consent.
4. 4. Biomedical research involving human subjects cannot legitimately be carried out unless the importance of the objective is in proportion to the inherent risk to the subject.
5. Every biomedical research project involving human subjects should be preceded by careful assessment of predictable risks in comparison with foreseeable benefits to the subject or to others. Concern for the interests of the subject must always prevail over the interests of science and society.
6. The right of the research subject to safeguard his or her integrity must always be respected. Every precaution should be taken to respect the privacy of the subject and to minimize the impact of the study on the subject's physical and mental integrity and on the personality of the subject.
7. Physicians should abstain from engaging in research projects involving human subjects unless they are satisfied that the hazards involved are believed to be predictable. Physicians should cease any investigation if the hazards are found to outweigh the potential benefits.
8. In publication of the results of his or her research, the physician is obliged to preserve the accuracy of the results. Reports of experimentation not in accordance with the principles laid down in this Declaration should not be accepted for publication.
9. In any research on human beings, each potential subject must be adequately informed of the aims, methods, anticipated benefits and potential hazards of the study and the discomfort it may entail. He or she should be informed that he or she is at liberty to abstain from participation in the study and that he or she is free to withdraw his or her consent to participation at any time. The physician should then obtain the subject's freely-given informed consent, preferably in writing.
10. When obtaining informed consent for the research project the physician should be particularly cautious if the subject is in a dependent relationship to him or her or may consent under duress. In that case the informed consent should be obtained by a physician who is not engaged in the investigation and who is completely independent of this official relationship.
11. In case of legal incompetence, informed consent should be obtained from the legal guardian in accordance with national legislation. Where physical or mental incapacity makes it impossible to obtain informed consent, or when the subject is a minor, permission from the responsible relative replaces that of the subject in accordance with national legislation. Whenever the minor child is in fact able to give a consent, the minor's consent must be obtained in addition to the consent of the minor's legal guardian.
12. The research protocol should always contain a statement of the ethical considerations involved and should indicate that the principles enunciated in the present Declaration are complied with.

II. MEDICAL RESEARCH COMBINED WITH PROFESSIONAL CARE (Clinical Research)

1. In the treatment of the sick person, the physician must be free to use a new diagnostic and therapeutic measure, if in his or her judgement it offers hope of saving life, re-establishing health or alleviating suffering.
2. The potential benefits, hazards and discomfort of a new method should be weighed against the advantages of the best current diagnostic and therapeutic methods.
3. In any medical study, every patient - including those of a control group, if any - should be assured of the best proven diagnostic and therapeutic method. This does not exclude the use of inert placebo in studies where no proven diagnostic or therapeutic method exists.
4. The refusal of the patient to participate in a study must never interfere with the physician-patient relationship.
5. If the physician considers it essential not to obtain informed consent, the specific reasons for this proposal should be stated in the experimental protocol for transmission to the independent committee (1, 2).
6. The physician can combine medical research with professional care, the objective being the acquisition of new medical knowledge, only to the extent that medical research is justified by its potential diagnostic or therapeutic value for the patient.

III. NON-THERAPEUTIC BIOMEDICAL RESEARCH INVOLVING HUMAN SUBJECTS (Non-Clinical Biomedical Research)

1. In the purely scientific application of medical research carried out on a human being, it is the duty of the physician to remain the protector of the life and health of that person on whom biomedical research is being carried out.
2. The subject should be volunteers - either healthy persons or patients for whom the experimental design is not related to the patient's illness.
3. The investigator or the investigating team should discontinue the research if in his/her or their judgement it may, if continued, be harmful to the individual.
4. In research on man, the interest of science and society should never take precedence over considerations related to the wellbeing of the subject.

APPENDIX 2 - DEFINITION OF ADVERSE EVENTS

Adverse Event

Any untoward medical occurrence in a patient or clinical trial subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment.

Comment:

An AE can therefore be any unfavourable and unintended sign (including abnormal laboratory findings), symptom or disease temporally associated with the use of an investigational medicinal product, whether or not related to the investigational medicinal product.

Adverse Reaction

All untoward and unintended responses to an IMP related to any dose administered.

Comment:

An AE judged by either the reporting Investigator or Sponsor as having causal relationship to the IMP qualifies as an AR. The expression reasonable causal relationship means to convey in general that there is evidence or argument to suggest a causal relationship.

Serious Adverse Event

Any untoward medical occurrence or effect that at any dose:

Results in death unrelated to original cancer

Is life-threatening*

Requires hospitalisation** or prolongation of existing inpatients' hospitalisation

Results in persistent or significant disability or incapacity

Is a congenital anomaly/birth defect

Or is otherwise considered medically significant by the Investigator***

Comments:

The term severe is often used to describe the intensity (severity) of a specific event. This is not the same as serious, which is based on patients/event outcome or action criteria.

* Life threatening in the definition of an SAE refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it were more severe.

**Hospitalisation is defined as an unplanned, formal inpatient admission, even if the hospitalisation is a precautionary measure for continued observation. Thus hospitalisation for protocol treatment (e.g. line insertion), elective procedures (unless brought forward because of worsening symptoms) or for social reasons (e.g. respite care) are not regarded as an SAE.

*** Medical judgment should be exercised in deciding whether an AE is serious in other situations. Important AEs that are not immediately life threatening or do not result in death or hospitalisation but may jeopardise the subject or may require intervention to prevent one of the other outcomes listed in the definition above, should be considered serious.

Serious Adverse Reaction

An Adverse Reaction which also meets the definition of a Serious Adverse Event.

Suspected Unexpected Serious Adverse Reaction

A SAR that is unexpected i.e. the nature, or severity of the event is not consistent with the applicable product information.

A SUSAR should meet the definition of an AR, UAR and SAR.

Unexpected Adverse Reaction

An AR, the nature or severity of which is not consistent with the applicable product information (e.g. Investigator Brochure for an unapproved IMP or (compendium of) Summary of Product Characteristics (SPC) for a licensed product).

When the outcome of an AR is not consistent with the applicable product information the AR should be considered unexpected.

APPENDIX 3 - COMMON TOXICITY CRITERIA GRADINGS

Toxicities will be recorded according to the Common Terminology Criteria for Adverse Events (CTCAE), version 4.0. The full CTCAE document is available on the National Cancer Institute (NCI) website, the following address was correct when this version of the protocol was approved:

http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm

APPENDIX 4 – MEASUREMENT OF MRD IN UKALL 2011

Therapy according to UKALL 2011 is stratified according to MRD as defined by real time quantitative PCR (RQ-PCR) analysis of Ig and TCR gene rearrangements at day 29 and upon recovery from consolidation at week 14 (Regimen C only). In addition MRD is measured at other time points as part of ongoing research within UKALL 2011.

MRD is measured by the laboratories of the UK MRD network acting as a virtual single laboratory. The RQ-PCR method is based on European best practice and in the developmental (pre NHS funding) phase has been subject to rigorous internal and external quality assurance.

Quality is overseen by the steering committee of the UK MRD network (Chair Professor Nick Cross) and the leukaemia subgroup of the NCRI Children's CSG.

Only MRD results generated by laboratories of the UK MRD network will be accepted for stratification of therapy in the UKALL 2011 trial. These are listed below.

Laboratories which are not currently members of the UK MRD network can apply to join. Applications should be made in writing to Dr John Moppett.

Acceptance will be at the discretion of the UK MRD network steering committee and inclusion will require fulfilment of **ALL** of the following criteria

1. Practical work to be carried out in a CPA accredited laboratory
2. Prior to adoption by the UK MRD network adequate performance in 3 sequential EuroMRD QA rounds.
3. Strict compliance with the UK MRD network SOP
4. Adequate performance defined according to UK MRD network during a 3 month "dry run" of production of results in real time for at least 30 patients. During this period the applicant lab will be required to self-fund staff and consumables.
5. All laboratories of the UK MRD network are required to be members of the EuroMRD QA network and to participate in the twice yearly quality control rounds and ongoing audit organised by the MRD network Operational Management Group.

Clinical centres opening this trial will agree to: -

Commission MRD services from a UK MRD network laboratory and work to a Service Level Agreement such that 90% of registered (and MRD eligible) patients can be stratified by MRD at day 29 and MRD data as agreed by the MRD network steering committee and Birmingham CRCTU is made available to the trial.

Nick Cross, chair of the NHS Molecular MRD Steering Committee can be contacted at: -

Nick Cross
Professor of Human Genetics
Wessex Regional Genetics Laboratory
University of Southampton
Salisbury District Hospital
Salisbury, SP2 8BJ, UK

Tel: +(44) 1722 429080
Fax: +(44) 1722 331531

E-mail: ncpc@soton.ac.uk

MRD LABORATORIES AND THEIR PRINCIPAL TREATMENT CENTRES

Adult centres wishing to enter patients into the trial should liaise with the local Children's Principal Treatment Centre (PTC) centre and laboratories listed below to send samples for MRD.

NHS Trust	Laboratory
Alder Hey Children's NHS Foundation Trust	Glasgow
Barts and The London NHS Trust	Bart's
Belfast Health and Social Care Trust	Glasgow
Birmingham Children's Hospital NHS Foundation Trust	Birmingham
Cambridge University Hospitals NHS Foundation Trust	Bart's
Cardiff and Vale University Health Board	Bristol
Central Manchester University Hospitals NHS Foundation Trust	Sheffield
Great Ormond Street Hospital for Children NHS Trust	Great Ormond Street
Heart of England NHS Foundation Trust	Birmingham
NHS Grampian	Glasgow
NHS Greater Glasgow & Clyde	Glasgow
NHS Lothian	Glasgow
Nottingham University Hospitals NHS Trust	Sheffield
Our Lady's Children's Hospital, Dublin	Glasgow
Oxford Radcliffe Hospitals NHS Trust	Bristol
Sheffield Children's NHS Foundation Trust	Sheffield
Sheffield Teaching Hospitals NHS Foundation Trust	Sheffield
Southampton University Hospitals NHS Trust	Bristol
The Christie NHS Foundation Trust	Sheffield
The Leeds Teaching Hospitals NHS Trust	Bristol
The Newcastle Upon Tyne Hospitals NHS Foundation Trust	Glasgow
The Royal Marsden NHS Foundation Trust	Bart's
University College London Hospitals NHS Foundation Trust	Great Ormond Street
University Hospitals Birmingham NHS Foundation Trust	Birmingham
University Hospitals Bristol NHS Foundation Trust	Bristol
University Hospitals of Leicester NHS Trust	Sheffield
University Hospital of North Staffordshire NHS Trust	Birmingham

CONTACT DETAILS FOR MRD LABORATORIES

Laboratory	Personnel	Contact Details
Bart's	Shaun Bevan Helen Warwicker Emma Burt	Barts Health NHS Trust Royal London Hospital Department of Molecular Haematology Room 201, 2nd Floor Pathology and Pharmacy Building 80 Newark Street Whitechapel London E1 2ES Tel: 0203 246 0142 Fax: 0203 246 0121 Shaun.bevan@bartshealth.nhs.uk helen.warwicker@bartshealth.nhs.uk suzanne.mcelwaine@bartshealth.nhs.uk emma.burt@bartshealth.nhs.uk
Birmingham	Jane Bryon Kerry Wall Max Rindl Johanna Hunt	West Midlands Regional Genetics Laboratory Birmingham Women's Hospital Edgbaston Birmingham B15 2TG Tel: 0121 627 2710 Fax: 0121 627 2711 JANE.BRYON@bwnft.nhs.uk KERRY.WALL@bwnft.nhs.uk MAX.RINDL@bwnft.nhs.uk Johanna.Hunt@bwnft.nhs.uk
Bristol	Dr Jeremy Hancock Paul Archer	Bristol Genetics Laboratory Pathology Sciences Southmead Hospital North Bristol NHS Trust Bristol BS10 5NB Tel: 0117 41 46173 / 41 46187 Fax: 0117 41 46464 Jeremy.Hancock@nbt.nhs.uk Paul.Archer@nbt.nhs.uk
Glasgow	Sandra Chudleigh Mary Gardiner	Department of Molecular Diagnostics (MRD Group)

	Frances Fee Linda Smith	Level 2 New South Glasgow Lab Building Southern General Hospital 1345 Govan Road Glasgow G51 4TF Tel: 0141 354 9110 Fax: 0141 232 7983 Sandra.Chudleigh@ggc.scot.nhs.uk Mary.Gardiner@ggc.scot.nhs.uk Frances.fee@ggc.scot.nhs.uk Linda.smith2@ggc.scot.nhs.uk
Great Ormond Street	Gary Wright Lana Mhaldien	Haematology Department Bone Marrow Laboratory Level 2 Camelia Botnar Laboratories Great Ormond Street Hospital For Children NHS Trust London, WC1N 3JH Tel: 0207 405 9200 x5400 Fax: 0207 813 8410 ukall.mrd@gosh.nhs.uk gary.wright@gosh.nhs.uk maria.ronayne@gosh.nhs.uk
Sheffield	Khalid Tobal James Blackburn Mark Watson	Sheffield Diagnostic Genetics Service Sheffield Children's NHS Foundation Trust Western Bank Sheffield S10 2TH Tel: 0114 271 7016 Fax: 0114 275 0629 khalid.tobal@sch.nhs.uk James.blackburn@sch.nhs.uk mark.watson@sch.nhs.uk

APPENDIX 5 - TREATMENT MODIFICATIONS FOR DOWN SYNDROME PATIENTS

Background

In ALL 97, Down Syndrome (DS) patients had a significantly worse EFS compared to non-DS patients (48% vs 78%) primarily because of a higher risk of CR death (28% vs <5%) due to sepsis. The majority of deaths occurred in consolidation and DIs. The increased risk of non-relapse mortality has persisted in UKALL 2003 and is also evident in ongoing COG trials. EFS of DS patients is also worse in BFM (58%) and NOPHO (54%) studies but not due to increased incidence of septic deaths.

Details of incidence, causes and timing of non-relapse deaths in DS patients in UKALL 2003 as of October 2010 are as below

DS mortality – October 2010

Total entered:	74
5 year EFS:	63%
Relapse:	6
Non-relapse deaths:	13 (17%)
Male	10/52
Female	3/22
Median age	5 years
NCI SR/HR	7/41, 6/33
Induction	4 (6%) (1 Reg A, 2 Reg B, 1 Reg C) (1 – parainfluenza pneumonitis, 1 – NEC + sepsis, 1 – pseudomonas, 1 – pneumonia and haemetemesis)
CR:	8(15%)
Consolidation	1 (Reg B – aspergillus)
DI1	1 (Reg A – sepsis)
DI2	1 (Reg A –sepsis)
Maintenance	5 (56, 89, 100, 112 and 140 weeks)

Treatment modifications

In view of the high non-relapse mortality, the following modifications will apply to the treatment regimens for Down syndrome patients, including those with lymphoblastic lymphoma (LBL):

- Randomisation- DS patients will be eligible for registration to UKALL 2011 but not the high dose methotrexate and pulses randomisation (R2).

- Dexamethasone randomisation to the short course should be for a continuous 14 days in children < 10 years and as intermittent therapy (weeks 1 and 3 only) in children ≥ 10 years.
- Daunorubicin should be omitted from induction for DS NCI High Risk and LBL patients. They should all receive the same 3 drug induction as NCI Low Risk patients (i.e. Regimen A induction).
- However, for Down syndrome patients with NCI HR BCP ALL and all cases of T-ALL if the day 15 BM shows a slow early response in ALL or there is concern over slow resolution of bulk disease in LBL, then in the absence of established serious morbidity, switch to Regimen C induction.

Post-induction-treatment for DS patients

Post-induction, DS patients will not be eligible for the second randomisation (R2). Therefore no drugs that these patients receive after induction will be IMPs. The following treatment approach should be considered for such patients following induction:

- Post-induction therapy is based on NCI Risk category & MRD Risk. MRD Low Risk and MRD No Result patients who had a rapid early response at day 15 receive Regimen A/B consolidation, depending on NCI risk, followed by a SINGLE DI.
- MRD High Risk patients and MRD No Result patients who had a slow early response should receive Regimen C (in the absence of serious morbidity during induction) with a SINGLE DI.
- Only MRD risk (or in absence of MRD result, slow early responders) patients should receive vincristine and steroid pulses during maintenance i.e. treat as per maintenance arm C1. MRD low risk patients should not receive pulses during maintenance i.e. treat according to maintenance arm A2.
- Boys should stop maintenance therapy after two years same as girls.
- Supportive care as per induction for Regimen B and C Consolidation and all DI blocks.

Supportive care

The following supportive care is recommended:

During induction:

- Administer prophylactic antibiotics. Ciprofloxacin 10mg/kg twice daily is recommended but individual centres may wish to use alternatives based on local infection and resistance patterns in discussion with their microbiologists.
- Review 3 times a week if an out-patient.
- Treat all febrile neutropenia episodes as high risk if using risk stratified policy for admission/intravenous antibiotics.
- DS patients may not present with classic signs of sepsis such as pyrexia. Treat non-specifically unwell patients as septic until proven otherwise.
- Be alert to early signs of shock in septic patients and refer promptly for intensive care.

APPENDIX 6 - DRUG INTERACTIONS WITH CYTOTOXIC AGENTS

These guidelines aim to address the more commonly reported and potentially serious drug interactions that may be encountered when using this protocol. **This should not be used as an exclusive list and current versions of SmPCs should be referred to wherever possible.**

These guidelines are not intended to be a comprehensive list of all potential drug interactions and normal precautions should be taken when prescribing any combination therapy.

INTERACTIONS WITH POTENTIALLY SERIOUS CLINICAL CONSEQUENCES

DRUG	POTENTIAL INTERACTION	COMMENTS
Vincristine	Itraconazole, voriconazole, posaconazole	Causes inhibition of the cytochrome P450 3A4 enzyme system resulting in increased incidence of peripheral neuropathy. ^{1,2} Hyponatraemia associated with SIADH has been reported with concomitant use of vincristine and azole anti-fungal agents ² RECOMMENDATION: AVOID USE OF THESE AZOLE ANTI-FUNGALS WITHIN ONE WEEK OF VINCRISTINE IF POSSIBLE. IF USED SUSPEND 48 HRS BEFORE AND AFTER VINCRISTINE DOSE.
	+/- Nifedipine	Nifedipine has been reported to enhance these effects.
	Fluconazole	Fluconazole is not reported to have the same adverse effects as other azole anti-fungals although could be predicted to have similar adverse effects
Anti-convulsants including phenytoin, carbamazepine, phenobarbitone	Possible reduced chemotherapy efficacy	An association between increased risk of relapse in children with B-lineage ALL and concomitant treatment with anticonvulsant therapy has been reported (EFS hazards ratio 2.67 95% CI 1.5-4.76, p=0.0009) ³ Many anti-convulsants induce hepatic enzyme activity. Significantly increased clearances of methotrexate and teniposide in patients receiving anti-convulsant therapy have been reported. Also see Phenytoin/Dexamethasone interaction.

Phenytoin	Dexamethasone	Phenytoin and dexamethasone mutually lower the efficacy of the other drug: Phenytoin increases hepatic enzyme metabolism of dexamethasone and lowered levels of phenytoin are reported with concomitant dexamethasone therapy RECOMMENDATION: AVOID THE USE OF PHENYTOIN, CARBAMAZEPINE AND PHENOBARBITONE IF POSSIBLE. POSSIBLE ALTERNATIVE ANTI-CONVULSANTS INCLUDE GABAPENTIN WHICH IS RENALLY EXCRETED AND DOES NOT INDUCE HEPATIC ENZYMES. CLONAZAPAM, CLOBAZAM, TOPIRAMATE OR LEVITIRACETAM HAVE NO KNOWN CLINICALLY RELEVANT INTERACTIONS WITH CYTOTOXIC DRUGS NB. VALPROATE HEPATOTOXICITY IS REPORTED AND SHOULD BE AVOIDED.
Combined Contraceptives (e.g. pills, patches and rings)	Increased risk of thrombo-embolic event	Patients using combined contraceptives should stop and switch to a low dose progesterone-only preparation, or norethisterone.
Intermediate / High dose methotrexate	Co-trimoxazole	Increased risk of methotrexate toxicity. Co-trimoxazole must be stopped at least 6 days prior to high dose MTX therapy in Protocol M/ M-A.
	Nephrotoxic drugs	Increased risk of nephrotoxicity resulting in delayed MTX excretion and increased risk MTX toxicity. Avoid all nephrotoxic drugs during Protocol M/ M-A (including aminoglycosides).
	Ciprofloxacin	May increase plasma levels of MTX leading to increased MTX toxicity. Avoid concurrent ciprofloxacin during Protocol M/MA
	Nitrous Oxide	Increases antifolate effect of methotrexate. Avoid concomitant use.

See next page for other potential interactions which require close monitoring.

OTHER POTENTIAL INTERACTIONS WHICH REQUIRE CLOSE MONITORING

DRUG	POTENTIAL INTERACTION	COMMENTS
Anti-coagulants Warfarin	Concurrent chemotherapy, especially 6-MP and steroids, INCREASES INR Co-trimoxazole DECREASES INR	The use of low molecular weight heparin for prophylactic therapy post thrombus formation would be preferable.
Mercaptopurine	Allopurinol	Avoid concurrent use. Can cause a 5 fold increase in AUC of 6-MP
Methotrexate as Capizzi methotrexate	Non Steroidal Anti-Inflammatory Drugs (NSAIDs) (including COX II inhibitors)	Increase in methotrexate levels due to competition for excretory pathways RECOMMENDATION: AVOID DURING CAPIZZI MAINTENANCE NB NSAIDs have adverse effects on platelet function
	Penicillins, Co-Amoxiclav	Penicillins reduce methotrexate excretion. RECOMMENDATION: AVOID DURING CAPIZZI MAINTENANCE NB Clavulenic acid is hepatotoxic
	Tetracyclines	Increased methotrexate toxicity through displacement of methotrexate from plasma binding sites NB Tetracyclines should be avoided in children under 7 years due to discolouration of teeth
High dose methotrexate	Non Steroidal Anti-Inflammatory Drugs (NSAIDs) (including COX II inhibitors)	Increase in methotrexate levels due to competition for excretory pathways RECOMMENDATION: AVOID DURING HIGH DOSE METHOTREXATE NB NSAIDs have adverse effects on platelet function Contd. on next page

High dose methotrexate (contd.)	Omeprazole	Possibly reduces excretion of MTX. Increased risk of MTX toxicity. Avoid concurrent use.
	Penicillins, aminoglycosides Co-Amoxiclav	Penicillins reduce methotrexate excretion. RECOMMENDATION: AVOID HIGH DOSE METHOTREXATE NB Clavulenic acid is hepatotoxic
	Tetracyclines	Increased methotrexate toxicity through displacement of methotrexate from plasma binding sites NB Tetracyclines should be avoided in children under 7 years due to discolouration of teeth
	Co-trimoxazole	Increased risk of methotrexate toxicity. Co-trimoxazole must be stopped at least 6 days prior to high dose MTX therapy in Protocol M/ M-A.
	Nephrotoxic drugs	Increased risk of nephrotoxicity resulting in delayed MTX excretion and increased risk MTX toxicity. Avoid all nephrotoxic drugs from 48 hours prior to administration until MTX has cleared during Protocol M/ M-A e.g. • Penicillins • Salicylates • NSAIDs including Cox II inhibitors • Probenecid • Oseltamivir • Aminoglycoside antibiotics • Vancomycin • Ciclosporin • Aciclovir
	Ciprofloxacin	May increase plasma levels of MTX leading to increased MTX toxicity. Avoid concurrent ciprofloxacin during Protocol M/MA.
Cytarabine	Flucytosine	Uptake of flucytosine by fungi may be inhibited by cytarabine
Cyclophosphamide	Suxamethonium	Duration and effect of neuromuscular blockade may be increased.

APPENDIX 7 - ADMINISTRATION OF HIGH DOSE METHOTREXATE

This appendix is intended to guide the safe administration of high dose methotrexate described during interim maintenance. It should be read in conjunction with the appropriate sections of the main protocol (Regimen A section 7.5.6, Regimen B section 7.6.6, Regimen C section 7.7.6)

Principles of High Dose Methotrexate Administration: In theory the use of high doses of methotrexate may be an advantage in lymphoblastic malignancy for two reasons: First, it may lead to increased uptake of methotrexate in resistant lymphoblasts and second it may lead to increased penetration of the drug to "sanctuary sites" such as the CNS and testis. However the administration of high dose MTX carries significant toxicity. This can largely be reversed by effective rescue within 42 hours of exposure. Failure to rescue a patient appropriately can lead to severe irreversible toxicity. Consequently rigorous attention to hydration, alkalisation, folinic acid rescue and serum methotrexate level monitoring are integral to the safe use of high dose methotrexate in UKALL 2011.

PREHYDRATION

Timing: Start hydration at least 6 hours prior to the commencement of the intravenous methotrexate.

Fluid: Dextrose/saline infusion fluid, bearing in mind the total sodium content. To each 500ml add 25mmol of sodium bicarbonate and 10mmol of potassium chloride. Alternatively sodium bicarbonate solution can be Y-sited to the hydration fluid.

Infusion rate: 125ml/m²/hr (3L/m²/day).

NB. Adjust the sodium bicarbonate concentration to maintain the urinary pH between 7 and 8. Do not start the infusion until a urinary pH of at least 7 has been achieved.

METHOTREXATE INFUSION AND HYDRATION

Administration of methotrexate

Dilute the methotrexate in saline (0.9%) or glucose (5%). Infuse 500mg/m² of methotrexate over 30 minutes then 4.5grams/m² of methotrexate to be infused over 23.5 hours. **Note, even if the infusion is not complete at this time point, it must be stopped.**

Hydration during methotrexate infusion

Fluid: Dextrose/saline infusion fluid, bearing in mind the total sodium content. To each 500ml add 25mmol of sodium bicarbonate and 10mmol of potassium chloride.

Infusion rate: Hydration needs to continue during 24 hours of methotrexate infusion to maintain a combined infusion rate of 125ml/m²/hr. This may be achieved either by using a Y extension set or using both lumens of the central venous line.

POST-METHOTREXATE HYDRATION

Continue hydration until folinic acid rescue is completed.

Fluid: Dextrose/saline infusion fluid, bearing in mind the total sodium content. To each 500ml add 25mmol of sodium bicarbonate and 10mmol of potassium chloride.

Infusion rate: 125ml/m²/hr (3L/m²/day).

FOLINIC ACID (LEUCOVORIN) RESCUE

It is important to note that for this schedule the folinic acid rescue starts very late and is short (3 doses in cases of normal methotrexate excretion). **Therefore the folinic acid should be administered intravenously, so that the rescue is optimal.**

Methotrexate plasma concentrations are measured at hours 24, (36*), 42, 48, (54*) hours from start of the methotrexate infusion.

*The 36 and 54 hour levels are only required if impaired methotrexate excretion is expected (see below).

The expected methotrexate plasma concentrations are given in the following table.

Time from start of methotrexate	Serum creatinine measurement	Methotrexate-level measurement	Methotrexate-plasma concentration expected	Folinic acid dose
24 hours	Yes	Yes	<150µmol/L	
36 hours	(yes)*	(yes)*	<3µmol/L	
42 hours		Yes	≤1µmol/L	15mg/m ² intravenous
48 hours	Yes	Yes	≤0.4µmol/L	15mg/m ² intravenous
54 hours		(yes)*		15mg/m ² intravenous

NB. Take care in interpreting results which are taken at different time points and processed by the lab at the same time.

FOLINIC ACID RESCUE REGIMEN WHEN LEVELS ARE WITHIN EXPECTED RANGE

1. If the 24 hour MTX level is <150µmol/L then folinic acid rescue begins 42 hours from the start of methotrexate infusion with the administration of a dose of 15mg/m² folinic acid.
2. If the 42 and 48 hour levels are ≤1µmol/L and ≤0.4µmol/L respectively then 15mg/m² folinic acid should be given at 42, 48 and 54 hours only as shown in the table below. No further folinic acid is required and no further levels are required

FOLINIC ACID RESCUE REGIMEN WHEN LEVELS ARE OUTWITH EXPECTED RANGE

1. If the 24 hour MTX level exceeds 150µmol/L or the creatinine increases to more than 125% of baseline, then increase hydration to 200ml/ m² per hour: - keep urinary pH >7.0. Estimate urine output 4 hourly and if urine output less than 80% of fluid intake consider Glucarpidase. Check 36 hour level
2. If the 36 hour level exceeds 10µmol/L then consider Glucarpidase regardless of urine output
3. If the 36 hour level is <10µmol/L then continue hydration to 200ml/m² per hour: - keep urine pH >7.0. Estimate urine output 4 hourly and if urine output less than 80% of fluid intake consider Glucarpidase. Check 42 hour level
4. If the plasma methotrexate concentration at hours 42, 48 from the start of methotrexate exceeds the normal range for time, increase the folinic acid rescue according to the schedule below.
5. If the plasma methotrexate concentration at hour 36 or 42 was high but then the patient "catches up", and the level falls to ≤1 µmol/L and ≤0.4µmol/L at 42 or 48 hours respectively then resume standard rescue as long as the urine output is satisfactory

(36 hour MTX level)	42 hour MTX level	48hour MTX level	Folinic acid rescue
Only required if 24 hr level is 150 μ M See below	1 to 9.9 μ mol/l	0.4 to 5.9 μ mol/l	Continue 15mg/m ² every 6 hours until MTX <0.1 μ mol/l (check level every 12 hours)
	10 to 39.9 μ mol/l	6 to 19.9 μ mol/l	Increase to 15mg/m ² every 3 hours until MTX <0.1 μ mol/l (check level every 6 hours) Consider Glucarpidase
	40 to 200 μ mol/l	20 to 100 μ mol/l	Increase to 100mg/m ² every 6 hours until MTX <0.1 μ mol/l (check level every 6 hours) Consider Glucarpidase
	>200 μ mol/l	>100 μ mol/l	Increase to 1000mg/m ² every 6 hours until MTX <0.1 μ mol/l (check level every 6 hours) Consider Glucarpidase

GLUCARPIDASE (Carboxypeptidase G2, Voraxaze®)

These are suggested guidelines only for the use of glucarpidase in case of methotrexate-induced renal failure and subsequent delayed methotrexate excretion following high dose methotrexate.

Glucarpidase is a recombinant glutamate carboxypeptidase that hydrolyses methotrexate (MTX) to inactive metabolites.

Glucarpidase could be considered when:

- as indicated in the table above
- serum creatinine rises by $\geq 100\%$ within 24 hours of MTX administration
- delayed excretion when plasma MTX levels plateau

Route

Intravenous

Dose

50units/kg as a single dose (all ages). Consider using it >10 μ mol/L at T=48hr (or according to established local practice).

Administration

Slow intravenous injection over 3-5 minutes. See SmPC for reconstitution details.

Stop folinic acid 4 hours prior to administration of glucarpidase and reintroduce 4 hours after treatment to replenish intracellular folate. The folinic acid dose should be based upon the pre-glucarpidase MTX plasma level.

Pharmacokinetics

Plasma MTX levels should decrease by >98% in 15 minutes. Elimination takes 8 hours. The DAMPA metabolite is known to cross-react with MTX in non-HPLC analysis of MTX levels resulting in an overestimate of plasma MTX levels. HPLC assays should be used if possible. Continue monitoring until levels decrease to <0.25 μ mol/L.

Repeat doses of glucarpidase carry a high risk of allergy or lack of efficacy due to tolerance. Hypersensitivity reactions are rare with the first dose.

Formulation & supply

1000units/vial; 2 vials per pack.

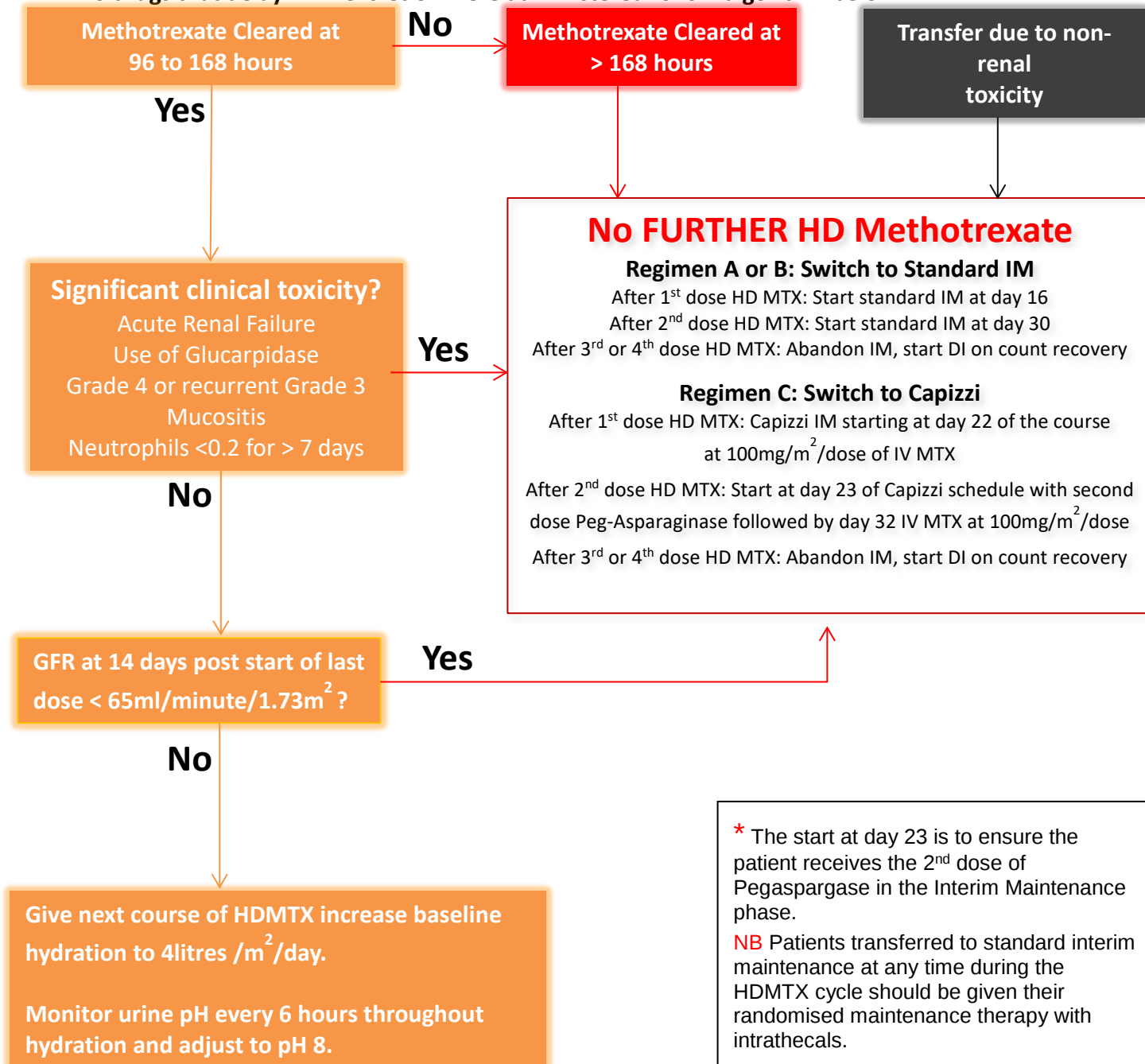
The sole supplier in the UK is Protherics (part of BTG). See Pharmacy Manual for details of ordering.

Methotrexate Excretion Delayed > 96 hours

Check previous concurrent interacting drugs with HDMTX. (See appendix 6, UKALL Protocol)

If any listed drugs were given, consider stopping these and re-expose to HD MTX if GFR normal.

If no drugs that delay MTX excretion were administered follow algorithm below:



APPENDIX 8 - MANAGEMENT OF ENCEPHALOPATHY RELATED TO INTRATHECAL METHOTREXATE

Background

Up to October 2010 215 cases of encephalopathy related to intrathecal methotrexate had been reported in patients entered on UKALL 2003. Typical presentation is with focal neurological deficit or rapid personality change or loss of consciousness occurring within 1 – 21 days (average 3 days) of exposure to intrathecal methotrexate (MTX). Full recovery was seen in 197 cases and in most cases this occurred within 48 hrs of the episode. In 15 cases patients had minor ongoing neurologic sequelae and in 3 cases methotrexate encephalopathy was thought to have played a major part in the patient's death.

Although episodes have occurred in all courses, just over half have been during the consolidation phase of Regimens B/C or part 2 of the Delayed Intensifications. This raises the possibility of an interaction with cytarabine in some patients. After a number of discussions as to the optimal approach to this condition the leukaemia sub-group of the paediatric oncology CSG have approved the following recommendations for managing such patients:

Management

Encephalopathy during Regimen B/C consolidation or part 2 of Delayed Intensification on Regimen C.

No further intrathecal MTX should be administered while patient is also receiving cytarabine (including in future courses containing cytarabine). These patients should be re-exposed, and in the absence of recurrence, administer missed doses during interim maintenance or maintenance. In the event of recurrence, change to intrathecal cytarabine + hydrocortisone in the doses given below. **These patients should not be randomised to high dose methotrexate.**

Encephalopathy during Protocol M

Discuss with the trial coordinators

Encephalopathy during other courses.

Re-expose as above and continue intrathecal MTX if no recurrence. Change to intrathecal cytarabine and hydrocortisone if problem recurs. Doses as below.

Doses:

Age	Ara- C	Hydrocortisone
1	20 mg	7.5 mg
2	25 mg	10 mg
3+	30 mg	12.5 mg

APPENDIX 9 - GUIDELINES FOR THE INVESTIGATION AND MANAGEMENT OF THROMBO-EMBOLIC EVENTS

Background

Thrombosis is a recognised complication of the treatment and management of ALL. The true prevalence in this patient population is unknown and varies with the method of assessment. The PARKAA study (Cancer 2003, Vol.97, 2) reported a prevalence of symptomatic events of 5% and asymptomatic events of 36.5% in children undergoing induction chemotherapy for ALL with a central venous catheter in situ. Asymptomatic events were diagnosed by screening with bilateral venography or MRI, echocardiography and a MRI of the head on completion of induction chemotherapy. Whilst the authors recommend carefully designed clinical trials of primary prophylaxis for the prevention of thrombo-embolic events in this patient population, there is at present insufficient data from children treated on UK protocols to support this. However, the need to collect such information to judge the appropriateness of prophylaxis is recognised. In general, primary prophylaxis for children with CVL cannot be recommended at this time, because there is no evidence for the efficiency or safety of this approach.

Asparaginase therapy and the presence of a central venous catheter are accepted as the main predisposing factors. The literature on the role of inherited thrombophilia in predisposing to thrombotic events in these children is conflicting, with the BFM reporting a significant association, and the Canadians and other major groups, no association. It is, therefore, considered premature to recommend universal screening or primary prophylaxis, although it may be prudent to try to identify less common high-risk abnormalities eg AT deficiency, PC deficiency.

The procedures described below are recommendations, however sites may continue to use local established practices.

Screening

- 1 Universal thrombophilia screening is not recommended.
- 2 A careful family history of thrombosis should be taken. Patients with a first or second degree relative with Protein C or A.T. deficiency should be screened for the relevant deficiency, as prophylaxis may be indicated. The literature is inconclusive for risk conferred by other inherited thrombophilias.
- 3 Girls and young women using combined contraceptives (including oral pills, topical patches and vaginal rings) should stop and change to a low dose progesterone-only preparation, or norethisterone.

Catheter (CVL) Related Thrombosis

1 Loss of CVL patency

Inability to withdraw blood +/- inability or impaired ability to infuse

- If there is no evidence to suggest displacement of the catheter tip and there are no signs to suggest the presence of occlusive thrombosis, proceed with urokinase (UK) lock
- Procedure: urokinase 2,500iu each lumen for 2-4 hours.

2 Failure to restore patency or recurrent loss of patency

- CXR to check position of catheter tip

- Linogram to assess CVL patency
- If the linogram demonstrates the presence of a fibrin sheath with no evidence of significant clot formation around the tip, proceed to urokinase infusion.
- Procedure: urokinase 150 iu/kg/hr via each lumen for 12-24 hours.
Monitor coagulation prior to and every 8 hours during infusion
- Low dose alteplase (0.1mg/kg/hr for 4 – 6 hrs) may be considered as an alternative thrombolytic agent
- Repeat linogram following infusion to confirm resolution.
- If the linogram demonstrates (or is suspicious of) the presence of significant clot formation around the catheter tip, or of vessel thrombosis, proceed with further imaging studies.
- Imaging: consider Doppler or MR venography.
- Comments on imaging –
 - Linograms have been shown to be relatively insensitive for the detection of large vessel CVL related thrombosis. In the presence of persistent line dysfunction despite a normal linogram, further imaging is indicated.
 - Doppler is a sensitive technique for imaging jugular veins, but has poor sensitivity for central intrathoracic veins.
 - MRV is less well evaluated but is likely to provide good sensitivity. Some patients are likely to require a GA for this technique.

3. Clinical symptoms/signs of CVL related thrombosis

- Arrange imaging to confirm the presence and extent of thrombosis.
- Imaging: consider Doppler, venography or MR venography.
- Complete thrombosis toxicity form

4. Doppler or Venography confirms the presence of large vessel thrombosis.

- Complete thrombosis toxicity form
- If the CVL is no longer required or is non-functioning it should be removed. If CVL access is required and the CVL is still functioning then the CVL can remain in situ.
- Unless otherwise contraindicated, anticoagulant therapy should be commenced.
- Comments on treatment –
 - Low molecular weight heparin (LMWH) is probably the anticoagulant of choice for initial therapy in most cases. Discussion regarding the most appropriate product and dosing with a local expert in paediatric haemostasis is recommended.
 - Prior to a lumbar puncture, or any other invasive procedure, the preceding two doses of LMWH should be omitted.

-
- If there is occlusive thrombus in a major vessel e.g. IVC, consider local thrombolytic therapy prior to anticoagulation and/or catheter removal. Low-dose alteplase (0.1mg/kg/hr) may be administered locally via the CVL but higher doses (0.5mg/kg/hr) are required for systemic therapy. Alteplase should be administered for 4 – 6 hours, followed by re-imaging.
 - Following the initial 3 months of therapy for patients with a first CVL-related DVT, prophylactic doses of oral anticoagulants (INR 1.5 to 1.8) or LMWH –(anti-factor Xa levels of 0.1-0.3) is an option until the CVL is removed. Patients with recurrent CVL related DVT should have prophylactic anticoagulation until the removal of the CVL.
 - Some patients will be scheduled to receive asparaginase as per protocol having had an earlier catheter-related thrombotic event. Consideration should be given to removal of the CVL but those patients receiving asparaginase with a CVL in-situ should receive prophylactic anticoagulation for the duration of the asparagine depletion.
 - Thrombophilia screening should be performed following completion of anticoagulant therapy and should include Protein C, Protein S, AT, FV Leiden, lupus screen, anticardiolipin antibodies and prothrombin gene 20210A.

CNS Thrombosis

Use of anticoagulants for treatment of the acute phase is contentious. Asparaginase should be suspended from that particular course but can be given in subsequent courses under prophylactic anticoagulant cover as described above Further detail is provided in Quereshi BJH 2010

APPENDIX 10 - TESTICULAR RADIOTHERAPY GUIDELINES

(Patients with testicular disease at diagnosis)

Standard treatment for those patients who still have a clinically enlarged testis at week 8 is testicular radiotherapy.

- a) Megavoltage or Orthovoltage apparatus may be used.
- b) As in previous MRC studies, the volume should include the testes and the spermatic cord to the level of the deep inguinal ring with lead shielding to surrounding tissues including the penis.
- c) The dose will be 24 Gy in 12 daily fractions of 2 Gy. This should be given during the first cycle of maintenance.
- d) Haemoglobin should be kept > 10 g/dL

APPENDIX 11– HEALTH ECONOMICS

Modern therapy for ALL is expensive: standard therapy as per UKALL 2003 or another international protocol with equivalent results costs approximately £50,000 per patient. Salvage therapy is much more expensive costing up to £200,000 per patient rescued. Thus in the absence of a trial the NHS will spend £28 million per annum on standard therapy for ALL in patients eligible for UKALL 2011. Little is known of the social cost of ALL therapy – specifically its impact on very long term health, family finance, academic and employment prospects as well as interpersonal relationships. UKALL 2011 is designed to further improve survival in ALL but significant emphasis is also placed on reduction of toxicity and a better balance between efficacy and cost. To this end a formal health economic analysis of UKALL 2011 will be commissioned during the course of the trial. This will aim to provide a much clearer indication as to which approach to therapy provides the best balance between efficacy and burden of therapy to the patient, family and NHS. In the interim the following issues are most pertinent understanding the impact of UKALL 2011 on NHS costs.

In line with international experience UKALL 2003 provided unequivocal proof that clearance of MRD is the most important prognostic factor in patients receiving homogenous therapy for ALL. Indeed MRD directed stratification of therapy is now internationally accepted as standard of care. In UKALL 2003 the costs of MRD testing were initially charitably funded by LLR but transited to NHS funding in 2009-10. At £3,170 per patient the cost of MRD testing in UKALL 2011 is identical to that in UKALL 2003 and thus cost neutral. It is important to recognise that UKALL 2003 led to an 8% reduction in relapse risk by comparison with ALL 99. As relapse therapy costs approximately £100,000 per patient the excess costs of MRD testing as per UKALL 2003 are more than offset by the reduced cost of salvage therapy when compared with ALL 99.

Following UKALL 2003 and other international studies it can now be argued that, within NCI and MRD risk stratified groups, no patient benefits from a second augmented interim maintenance or any type of second delayed intensification. Thus, in UKALL 2011 standard therapy is arm A or B with a single DI in MRD low risk and Arm C with a single DI in MRD intermediate risk and LBL. This shortening of post-induction intensification represents a reduction in drug costs of £1500 per patient as well as a significant reduction in inpatient admissions by comparison with the standard 2 DI arms of UKALL 2003.

There is no excess drug cost associated with the induction dexamethasone randomisation. By contrast excess drug costs will be incurred by the second randomisation (R2). It is estimated that 80% of patients will be eligible for the methotrexate and pulses randomisations. Of these 40% will be randomised to protocol M at an excess drug cost of £720 per patient allocated high dose methotrexate. This will in part be offset by a saving of £350 per case in the 40% of patients randomised to not receive pulses. The excess in patient days associated with administration of Protocol M are offset by the reduction in day case admissions for intrathecal chemotherapy and will likely be further offset by reduced admission for central line associated infection in the no pulses arm.

Taken together the excess drug costs of the randomisations in UKALL 2011 are £29,600 per 100 patients enrolled or £130,200 per annum. This is offset by over £660,000 per annum saved from the adoption of one delayed intensification course for all patients. Moreover at 0.78 million pounds over the course of the trial, the excess drug costs of the randomisation represent less than 1% of the cost of delivering standard ALL therapy out with a trial. By implication the delivery of standard therapy alone precludes further progress toward a better understanding of the optimal balance between benefit and cost in ALL.

NHS trusts and PCTs may also find it more helpful to think of the excess cost in terms of the extra cost of treatment per patient entered into the trial. Since we expect 440 patients/ annum to be entered into the trial, for each patient entered into the trial, the excess service cost to the NHS will be £130,200 divided by 440 = £295. Lastly it is anticipated that UKALL 2011 will reduce the risk of CNS relapse from 4% to 2%. Currently, it costs up to £200,000 to successfully treat one child with CNS relapse of ALL as most will need a stem cell transplant. Even a 1% reduction in CNS relapse will lead to 4 fewer relapses/annum in the UK at an annual cost saving of £670,000.

APPENDIX 12 – ASPARAGINASE STUDY- CLOSED

The Asparaginase study closed to recruitment on 25th July 2017.

Aims

1. Determine the pharmacokinetics of dexamethasone and L-Asparaginase in childhood ALL induction therapy
 - (i) Assess pharmacokinetics of dexamethasone in each arm of the steroid randomisation in ALL 2011, in regimen A and B patients (n = 250)
 - (ii) Determine trough L-Asparaginase (ASNase) activity and anti-ASNase activity each arm of the steroid randomisation in ALL 2011, in regimen A and B patients (n = 800)
 - (iii) Compare drug levels with drug-specific toxicities
2. Investigate the role and effect of synergy between L-Asparaginase and dexamethasone
 - (i) Investigate dexamethasone and ASNase kinetics with risk factors, early response to therapy and MRD
 - (ii) Evaluate the synergy between Dexamethasone and ASNase and the effect when ASNase is given alone, on treatment response
 - (iii) Analyse the effect of dexamethasone kinetics in those with inadequate asparaginase activity in induction and delayed intensification
3. Correlate L-ASN scheduling and activity with methotrexate clearance and toxicity in interim maintenance of patients treated on Regimen C

Background

Our previous clinical trials have shown the superiority of dexamethasone (ALL 97/99) and the benefit of sustained L-Asparaginase activity (ALL 2003). There are now data to suggest that these two drugs are best used in conjunction, exhibiting a synergistic interaction. We have also shown that the high-risk cytogenetic subtype iAMP21 benefits from intensive schedules incorporating intravenous methotrexate (ALL 2003). However there is wide inter-individual variation in pharmacokinetics of all 3 drugs, and collectively they contribute significantly to toxicity in ALL patients. Side effects like sepsis, psychosis, encephalopathy and pancreatitis are potentially fatal, while others including avascular necrosis, thrombosis and diabetes lead to long-term disability. The new study ALL 2011 is designed to evaluate if we can reduce the steroid burden and target high dose therapy more effectively. We will take advantage of its design to investigate individual variations in pharmacokinetics, explore synergy and correlate this with early response to therapy, toxicity and outcome. Results generated will lead to future individualisation of therapy to decrease toxicity and further improve outcome.

Methodology

For the pharmacokinetic studies, blood samples will be collected on the study days shown in Figure 1. Blood samples (3ml) will be obtained pre-treatment and at 1, 2, 4 and 8h after the first dose of dexamethasone on days 1 and 14 (short arm) or 28 (standard arm) of treatment. All samples will be taken from a central venous line or peripheral cannula. Plasma samples will be obtained immediately following sample collection and stored at -20°C prior to transport to Newcastle for analysis. Diagnostic bone marrow samples will also be obtained. For the ASNase studies, all samples will be sent to the Leukaemia Cell bank for processing prior to batch analysis (approved by the cell bank committee). In addition to the tests described above, we will also look for anti PEG and anti ASNase antibodies in patients who show inadequate ASNase activity. The methodology is summarised in Table 1.

Table 1. Proposed Assays

Drug	Sample	Assay	Sensitivity
Dexamethasone	Plasma	LC/MS	0.05 ng/ml
ASNase	Plasma	Chromogenic	34u/L
Anti-ASNase	Plasma	ELISA	N/A
Genotype	DNA	PCR / deep sequencing	N/A
MVs	Plasma	Antigen capture	N/A
Thrombogenic assay	Cells	Phospholipid-dependent assay	N/A

Statistical Power and Pharmacokinetic Analysis

Power calculations are based upon a two group comparison of dexamethasone clearance, i.e. comparing standard versus short dexamethasone treatment. Clearance is assumed to be log-normally distributed with an SD of approximately 0.7³. Hence, total CV is approximately $\sqrt{0.46^2 + 0.53^2} = 0.70$. With a study population of 250 patients, the study would have >90% power to detect a 40% relative difference in dexamethasone clearance between the defined groups. Inclusion of a minimum of 50 younger children <5 years of age provides a 90% power to detect a 57% relative difference between younger and older patient cohorts. Comparable differences in dexamethasone clearance would be required to provide a similar study power for patients separated into two groups based on MRD risk status at day 28. Based on the predicted incidence of inadequate ASNase activity (14%), a study with 250 patients would have 90% power to detect a 50% difference in dexamethasone clearance between patients with adequate and inadequate ASNase activity.

Data obtained will be used to determine pharmacokinetic parameters including AUC, clearance and half-life for dexamethasone in each of the defined treatment groups. Pharmacokinetic modelling will be carried out using these data in conjunction with patient characteristics and clinical parameters in order to investigate key factors involved in determining individual drug exposures within the defined patient populations. The influence of pharmacokinetic parameters and genotype on day 28 MRD status, event free survival and grade III/IV toxicity will be investigated using Cox proportional hazards models and t-test analysis respectively.

Laboratory Assays

Asparaginase Activity: The diagnostic sample will serve as baseline, and subsequent samples are to be taken 7-14 days after the last pegaspargase and 1-1.5 days after crisantaspase. The quantification of enzymatic activity of all forms of asparaginase is based on the measurement of substrate turnover at a maximum rate. One unit of activity is defined as the amount of enzyme which releases 1mmol of ammonia and aspartate from 1mmol asparagine per minute at 37°C. The liberated ammonia can be measured spectrophotometrically either after nesslerization or by an enzyme-coupled reaction. A number of assays have been developed. Asparaginase activity in plasma samples is quantified by incubating the samples with an excess amount of L-aspartic acid β -hydroxamate (AHA) at 37°C. Asparaginase hydrolyses AHA to aspartic acid and hydroxylamine, which is detected at 710 nm after condensation with 8-hydroxyquinoline and oxidation to indooxine. This method requires 20ul of plasma and has a sensitivity of 30U of Asparaginase/L of plasma. The coefficients of variations for inter- and intra assay reproducibility are 10.8% and 7.1% respectively.

Asparaginase Antibody Assay: Briefly, microtitre plates are coated with purified (and recombinant) E coli asparaginase. The positive anti-asparaginase antibody controls, calibrators with defined anti-asparaginase reactivities, normal human serum as negative control, and patient serum samples at certain dilutions are added and incubated for 1 h. After washing, a polyclonal goat anti-human IgG and IgM horseradish peroxidase conjugate is added and incubated for 1 h. After washing, 3,3',5,5'-tetramethylbenzidine is added and incubated for 30 min. Anti-asparaginase antibody levels are measured at 450 nm for the enzymatic product (subtracting the absorbance at about 630 nm for nonspecific absorbance) using a microplate reader. The OD values of the calibrators are plotted

against their corresponding concentration, given as arbitrary units per ml, to construct a calibration curve over the whole measuring range of the assay. Positive reactivity in serum is calculated using this calibration curve.

Samples

All peripheral blood samples are collected in EDTA tubes at routine sampling times required by the protocol. No additional invasive tests are required.

All samples (peripheral blood) are for asparaginase activity and antibody detection. For this we need to know the date the last dose of asparaginase was given and the date the sample was taken. Please ensure that the peripheral blood sample is taken at least 7 days and not more than 14 days after the last dose of pegaspargase.

IF PATIENT IS ON CRISANTASPASE (ERWINIA L-ASPARAGINASE), PLEASE TAKE A BLOOD SAMPLE 24 HOURS AFTER A DOSE I.E. JUST BEFORE THE NEXT DOSE.

Please fill out an 'Asparaginase Study Form UKALL 2011' (which can be found in the Investigator Site File) with each sample and notify on the bottom of the form if there is hypersensitivity. Cell Bank sample packs should be used to collect and send the samples.

During induction

5mls of peripheral blood in EDTA tube on day 9 (along with 2nd Vincristine.)

5mls of peripheral blood in EDTA tube on day 16 (along with 3rd Vincristine and 12 days after the first asparaginase.

5ml of peripheral blood in EDTA tube on day 30 (along with the 5th Vincristine.)

During Delayed intensification

5ml of peripheral blood in EDTA tube on day 16 (along with the 3rd Vincristine)

During Maintenance therapy

10ml of peripheral blood in EDTA tube at any convenient time point during maintenance. Please note this sample will also be used for epitope analyses and therefore requires an adequate number of white cells. Please ensure that WCC >1.5 x 10⁹/L.

Additional samples for Regimen C

Augmented BFM Consolidation

5mls of peripheral blood in EDTA tube on any ONE occasion on either day 23 or day 51. Can coincide with Vincristine.

Capizzi

5ml of peripheral blood in EDTA tube on day 12

5ml of peripheral blood in EDTA tube on day 32 along with the 4th Vincristine.

Protocol M-A

5 ml of peripheral blood in EDTA tube on day 15.

5 ml of peripheral blood in EDTA tube on day 29.

For patients on crisantaspase (pegaspargase hypersensitivity): Patients who are on Crisantaspase due to pegaspargase hypersensitivity send 5ml of peripheral blood in EDTA tube 24 hours after the last dose of crisantaspase (trough levels) on one of the following days: day 4, 18, 32 or 46.

References

Lanvers C, Vieira Pinheiro JP, Hempel G et al. Analytical validation of a microplate reader-based method for the therapeutic drug monitoring of L-asparaginase in human serum. *Anal Biochem* 2002;309:117-26.

APPENDIX 13 – QUALITY OF LIFE STUDY

Aims:

1. To assess prospectively the health related quality of life (QoL) steroid related quality of life (QuEst) of the children and young people treated in the UKALL 2011 trial for acute lymphoblastic leukaemia and lymphoma.
2. To describe changes in QoL throughout treatment and up to the completion of therapy.
3. To compare the impact of the different treatment regimens in UKALL 2011 on QoL as follows:
 - (i) The use of higher dose, shorter regimen of dexamethasone during induction
 - (ii) the addition of intravenous methotrexate as CNS directed therapy
 - (iii) the impact of maintenance 'pulses' of vincristine and dexamethasone
4. To assess the 'burden of therapy' for the child and (where appropriate) their family throughout therapy.

Background

QoL may be explored in a health context in a number of ways¹. These include:

1. objective measures, such as clinical indices that patients would not themselves use or necessarily be aware of (eg fever, thrombocytopenia, mucositis)
2. functional performance (the ability to perform daily activities about which patients are aware, such as climbing stairs)
3. patient's own evaluation of the subjective experience of being able to complete a given activity

Attempts to determine QoL have included the use of proxy measures such as school absence or the use of related measures already available for children. These include, for example, measures of self-esteem or depression. Reliance on any of these individual measures is limited as none alone will provide a comprehensive or sensitive indicator of QoL during treatment for leukaemia. On the other hand the use of multiple measures or 'batteries' is cumbersome and they tend to be lengthy, repetitive and may lack sensitivity to detect the specific impact of cancer on the child's QoL. For this reason, a number of specifically developed measures of QoL for children with cancer have been reported. The measure chosen for this study for patients up to the age of 16 is the PedsQL by Varni which fulfils the following criteria:

1. Availability of parallel versions, enabling comparisons to be made between parent and child views about QoL.
2. Adequate psychometric properties. Varni et al² reported that children who had completed treatment had a better QoL than those who were still on treatment.
3. Brevity. The PedsQL remains one of the most brief and comprehensive measures of QoL.
4. Availability of population norms. Varni published norms for the US population and the properties of the measure in the British population have also been published³. Comparisons with norms will be made to enable us to distinguish changes in QoL which are the result of treatment compared with any that might be attributable to normal age-related changes.
5. Extensive use of the PedsQL measure in UKALL 2003, demonstrating excellent compliance from families and face validity in this context.

Preliminary data from UKALL 2003

The PedsQL questionnaire has been validated in UKALL 2003, in children. The assessment of QoL undertaken in UKALL 2003 has demonstrated that the measures used clearly show a change in QoL during the early stages of treatment in boys and girls. This is seen in all domains, with a fall in QoL scores early in treatment followed by some improvement over time. Analysis of the randomised groups has not yet been undertaken however, it is clear that the instrument used is sensitive to changes in QoL in this population over time. Below is an example of parental scores for the 5 domains (n = 235):

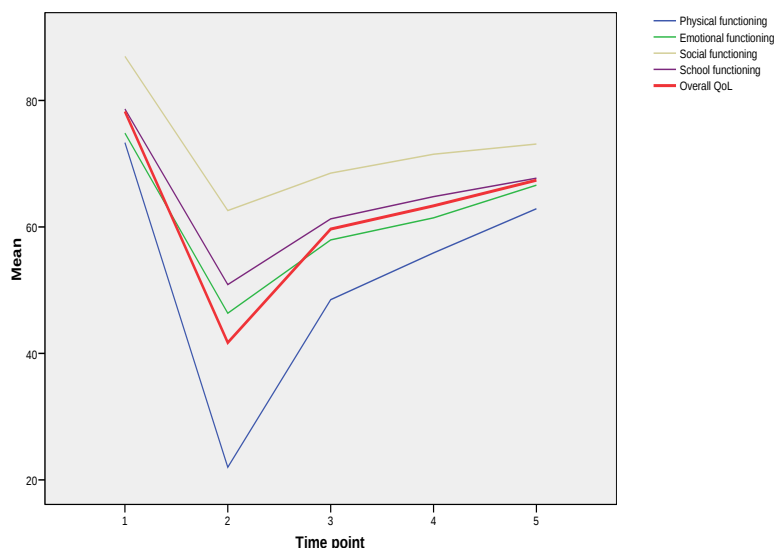


Figure 1 Plots of parental scores for all domains in 235 children treated according to UKALL 2003. Time points t1 - week 1; t2 - week 4; t3 - start of maintenance t4 - 18 months; t5 - end of treatment.

Detecting impact of steroids on QoL using QuEST

As stated above, two of the trial randomisations concern the use of dexamethasone. In addition to the well-documented physical side effects, corticosteroids are known to have adverse effects on mood, cognitive function and behaviour. It is therefore important that when analysing QoL during treatment that the measures used are sensitive to the effects of steroids. Within UKALL 2003, QoL assessment demonstrated a clear reduction in QoL scores during induction therapy with a gradual improvement over time (as noted above). However, there were no significant differences identified related to specific treatment interventions.

The Quality of Life Evaluation on Steroids (QuEST) tool has been developed as a treatment specific measure evaluating the impact of steroids on QoL. It has been developed in the UK with participation of patients from 4 UK centres undergoing maintenance therapy for ALL. Initial testing shows the measure to be both reliable and valid (6,7). The final validity assessment will take place following the completion of UKALL 2011.

The QuEST tool comprises 20 questions (taking 5-10 minutes to complete) for patients and parents and is formatted so that it can be used alongside PedsQL™. Responses will contribute to the overall QoL assessment. In addition, responses to the QuEST tool will be compared to those of the PedsQL™ in order to confirm construct validity. Responses to QuEST at different time points during maintenance therapy will also be used to perform a test-retest reliability assessment.

Family burden of care

Practical issues that affect day to day life are not included in contemporary QoL measures. A comprehensive evaluation needs to take into account parents' ability or willingness to carry out home-based treatments and care and the impact of the child's illness on the family. In the absence of any standardised measure, we previously devised a questionnaire based on clinical and research

experience. This questionnaire has been well received by families and will be further refined for UKALL 2011. The questionnaire has been integrated with the PedsQL and is detailed below.

Measurement of QoL in UKALL 2011

The UKALL 2011 design allows investigation of the impact of very different treatment regimens on the quality of life of patients and their families. Many variables may have an impact on the child's health related quality of life. This is particularly true during induction chemotherapy when the patient may be acutely unwell and is overcoming the shock of the diagnosis of a life threatening condition. QoL will be assessed at diagnosis and again at the end of induction allowing a comparison between the two different induction steroid regimens (short or standard dexamethasone dosing). During the maintenance phase of treatment, patients are in a more stable state and thus the trial provides a real opportunity to understand the impact of both the use intravenous methotrexate rather than continued intrathecal therapy and the impact of pulses of steroids and vincristine.

Assessment of QoL in 16-25 year olds

QoL questionnaires that are age-appropriate with established reliability and validity will be used. Generic QoL will be assessed using the SF-12 (4) and cancer specific QOL using the QLQ-C30 (5). In addition separate questions to determine participation in daily activities will parallel those used in the child study.

Method

Eligibility for QoL study:

1. Entered on UKALL 2011
2. No diagnosis of neurodevelopmental disorder (e.g. Down syndrome)
3. At least one parent able to speak/ read English

Questionnaire (QoL and QuEST) administration

Questionnaires will be completed during routine clinic visits. Questionnaires will be given to all parents of patients aged 3 to 15 years. They will also be given to all patients aged 8 years and over.

Please note:

1. All patients entering the trial at <3 years of age will reach this age at some point during treatment and questionnaires can be given to parents of these children at the designated time point after their birthday.
2. Those patients who reach the age of 8 years during treatment will be eligible to complete the questionnaires themselves from that time point, even though they have not previously done so.

Five assessments will be recorded for QoL and QuEST .

1. **During week 1.** Baseline assessment (No burden of care questionnaire at this time point). This questionnaire should ideally be completed after informed consent has been obtained and before the first randomisation/commencement of treatment. To be completed within first week at the latest.
2. **End of induction treatment.** This allows a direct comparison of the induction regimens. This questionnaire must be completed within 2 weeks of completing induction.
3. **At the end of interim maintenance.** This allows comparison of the immediate impact of intravenous methotrexate. This questionnaire must be completed within 2 weeks of the end of interim maintenance.
4. **At 18 months after registration.** This allows comparison of the impact of therapy on patients who have previously received methotrexate and those who are receiving pulses of dexamethasone and vincristine. This questionnaire must be completed within 2 weeks of this time point.
5. **End of treatment.** A final assessment at the completion of therapy, which will be at approximately 24 months for girls and 36 months for boys. Evaluation of the length of treatment on QoL and family burden of care with ongoing comparison between the groups randomised to pulses of vincristine and dexamethasone and those not receiving these. This questionnaire must be completed within 2 weeks of this time point.

Recruitment

The Quality of Life study is an integral part of the UKALL 2011 trial and thus patients will be recruited to the study at the time of registration to the UKALL 2011 trial. Questionnaires will be given by the treating clinician, a research nurse or nurse specialist within the unit, based on patient eligibility and age at the assessment time point. Parent questionnaires will be given to those with children aged 3 to 15 years inclusive. Patient questionnaires will be given to all those aged 8 or over. Most will be able to complete these with minimum explanation (time for completion approximately 10 minutes). Nurses will be given specific instructions about administration of questionnaires and ongoing support throughout the running of the study by the QoL leads.

Statistical design

The QoL study piloted in UKALL 2003 shows a drop of more than one standard deviation in the mean values for the three main measures (physical, emotional and social functioning) at the end of intensification, increasing back by 50-80% of normal by the end of treatment. Should this improvement occur earlier in the no-pulses arm, measurement at around 18 months will show a difference of 0.25-0.4 standard deviations (sd). To detect 0.25 SD would require 340 patients per treatment group to achieve 90% power from a crude comparison between randomised treatments at this time point, and 250 per group for 80% power.

The measurement of QoL using the PedsQL questionnaires for Children and SF 36 for young people is integral to UKALL 2011. It is anticipated that in addition a more 'in depth' study of QoL will be undertaken in some patients through the vincristine and dexamethasone pulses, together with the development of new tools to explore the economic and social burden of therapy on families and the cost of therapy to the NHS.

1. Gill, T. M. & Feinstein, A. R.: A critical appraisal of the quality of quality-of-life measurements. *Journal of American Medical Association*, 1994; 272: 619-626.
2. Varni, J., Burwinkle, T. M., Seid, M., and Skarr, D. (2003) The PedsQL™ 4.0 as a Pediatric Population Health Measure: Feasibility, Reliability, and Validity. *Academic Pediatrics*, 3, 6, 329-341
3. Upton P, Eiser C, Cheung I, Hutchings H, Jenney M, Maddocks A, Russell I, Williams J. Measurement properties of the UK-English version of the Pediatric Quality of Life Inventory™ 4.0 (PedsQL™) generic core scales. *Health and Quality of Life Outcomes* 2005, 3:22.
4. Ware JE, Kosinski M, Keller S, A 12 item, short form health survey: construction of scales and preliminary tests of reliability and validity. *Medical Care* 1996;34:220-233.
5. Aaronson NK, Ahmedzai S, Bergman B, Bullinger M, Cull A, Duez NJ, Filiberti A, Flechtner H, Fleishman SB, de Haes JC, et al. The European Organization for Research and Treatment of Cancer QLQ-C30: a quality-of-life instrument for use in international clinical trials in oncology. *J Natl Cancer Inst.* 1993 Mar 3;85(5):365-76.
6. Adams MR, Sherratt S, Johnson, Jenney MEM. Development of the EQuALL (Evaluating Quality of Life in ALL) questionnaire: a treatment-specific measure for the effects of dexamethasone during treatment for childhood acute lymphoblastic leukaemia. SIOP abstracts. *Pediatric Blood and Cancer (E-journal)* 2012
7. Adams M, Robling M, Grainger J, Tomlins J, Johnson A, Morris S, Velangi M, Jenney M. Quality of life Evaluation in patients receiving Steroids (the QuEst tool): initial development in children and young people with acute lymphoblastic leukaemia. *Arch Dis Child.* 2015 Dec 23. pii: archdischild-2015-309139. doi: 10.1136/archdischild-2015-309139. [Epub ahead of print]

APPENDIX 14 – FLOW MRD STUDY –CLOSED

The Flow MRD study closed to recruitment on 6th June 2016.

NB. This study is for Acute Lymphoblastic Leukaemia (ALL) patients only.

Sampling centres: Birmingham, Bristol, Cambridge, Glasgow, GOSH, Leeds, Liverpool, Manchester, Newcastle, Nottingham, Oxford, Southampton

Analysis centres: Birmingham, Bristol, Glasgow, GOSH, Newcastle

Background

Flow cytometric MRD analysis is cheaper and quicker than the current UK molecular method. There is now good evidence that flow analysis of MRD is predictive of relapse risk. Studies from COG, St Jude, and BFM group as well as UK studies performed at Birmingham Children's Hospital and in the UK Flow MRD Laboratory Network have shown the following;

- a) Flow MRD has high applicability (>90%) and adequate sensitivity (0.01%) to allow application to the vast majority of patients and to allow identification of both high risk and low risk patients (the latter particularly if used in conjunction with other risk factors). Applicability and sensitivity can be increased by the use of 6 or 8 colour flow and standardized approaches to this have now been published by the BFM and Euroflow consortia.
- b) The results of molecular and flow cytometric analysis of MRD are not interchangeable as they measure different things – DNA vs. live cells. Most studies have shown good concordance between the two methodologies but there are also discordant measurements. Concordance in UK studies is 82% with discordance occurring in both directions.
- c) Flow MRD status at the end of induction is predictive of outcome. As with molecular MRD, the relative risk of relapse in Flow MRD positive and negative groups is dependent on the treatment protocol and method used.
- d) Flow MRD at early time points (day 8 or 15 of treatment) may be able to rapidly identify BOTH patients with very high risk of relapse and those with very low risk of relapse – further analysis of data accrued during UKALL 2003 is needed to determine this further.

Proposed Study

Building on the expertise gained during UKALL 2003, this limited centre add on study will explore the predictive value of 6 colour flow cytometric assessment of MRD in UKALL 2011. The ultimate aim is to develop a flow cytometric system for MRD stratification that is as robust as the current molecular approach yet quicker, cheaper and more widely available.

Aims

1. Analysis at diagnosis – to further assess if any particular immunophenotype is predictive of prognosis
2. Assessment during induction therapy at all time points will allow comparison of disease kinetics in the 2 randomised dexamethasone arms and may identify different risk groups according to the pattern of disease response
3. Analysis at day 8 and day 15 of induction therapy

- (i) To assess identification of high risk patients. Analysis in UKALL 2003 showed that flow MRD at day 8 or 15 was predictive of outcome, but follow up was short and reanalysis will be performed every 6 months to see if this time point can be used for risk stratification
- (ii) To assess identification of very low risk patients. Analysis in UKALL 2003 showed that all patients with flow MRD <0.1% at day 8 or 15 have remained in complete remission (CR).
4. To perform functional studies throughout therapy.
5. Analysis at the end of induction and later time points to allow comparison with molecular results
6. Assessment of the overall predictive value of flow MRD analysis and comparison with molecular MRD analysis in a cohort of patients receiving standardised therapy
7. International comparison of MRD analysis with COG and IBFM Flow MRD groups

Sample Requirements

2-5ml marrow/peripheral blood in ACDA at the time points specified:

1. Day 0 to identify markers for MRD follow up (marrow – blood only required if poor marrow sample)
2. Day 8 /15 (paired marrow/peripheral blood)
3. Day 29 (marrow only)
4. Day 22 Consolidation (Week 9 - Regimen C).
5. Upon recovery from consolidation at week 14 for Regimen C patients (marrow only)

Dedicated samples will be sent from defined treatment centres to laboratories within the UK Flow MRD Network. Clinicians will be blinded to the results. The costs of this study will be funded by Leukaemia and Lymphoma Research.

Please send samples on day of collection by first class post.

Please email analysis centres to notify samples sent.

Sample size and power calculations

150 in each dexamethasone arm. 300 patients randomised equally between the 2 dexamethasone arms would give 90% power to rule out a difference of 14% from a standard arm rate of 80%, or 16% from a rate of 70%, allowing for 20 drop out.

Sample Distribution

Hospital Site to send MRD samples	Analysis Laboratory	Contact Details
Leeds Liverpool Manchester Newcastle Southampton	Newcastle	Newcastle Flow MRD Team Northern Institute for Cancer Research Paul O’Gorman Building Framlington Place Newcastle upon Tyne NE2 4HH Julie Irving/Maryna Sanichar ☎ 0191 246 4369 ✉: J.A.E.Irving@newcastle.ac.uk

Birmingham Cambridge Oxford	Birmingham	Birmingham Flow MRD Team Haematology Department Birmingham Children's Hospital NHS Foundation Trust Whittall Street Birmingham B4 6NH Jenny Jesson/Lisa Eyre ☎ 0121 333 9865/9868 ✉ jennifer.jesson@bch.nhs.uk lisa.eyre@bch.nhs.uk
Bristol Nottingham	Bristol	Mr Paul Virgo Department of Immunology Southmead Hospital Westbury-on Trym Bristol BS10 5NB ☎: 0117 959 6306 ✉: paul.virgo@nbt.nhs.uk
GOSH	GOSH	Sarah Inglott Chief BMS and section head Bone Marrow Lab Level 2, room P2.013 Haematology Camelia Botnar labs Great Ormond Street Hospital London WC1N 3JH ☎: 0207 829 7901 (direct line) ✉: inglos@gosh.nhs.uk
Glasgow	Glasgow	Linda Knotts Chief BMS Royal Hospital for Sick Children Haematology Department Dalnair Street Glasgow ☎: 0141 201 0388/9391 ✉: linda.knotts@ggc.scot.nhs.uk

APPENDIX 15 – DEXAMETHASONE PHARMACOKINETIC STUDY - CLOSED

The Dexamethasone Pharmacokinetic study closed to recruitment on 11th January 2018.

Aims

1. To investigate the potential for dexamethasone pharmacokinetic parameters to provide biomarkers for clinical response (day 29 MRD status and EFS) and incidence of grade III/IV steroid-related toxicity including myopathy, osteonecrosis and behavioural problems.
2. To determine the relationship between dexamethasone clearance and serum albumin concentration.
3. To evaluate potential synergy between dexamethasone and asparaginase.
4. To determine differences in dexamethasone pharmacokinetic parameters (predominantly AUC) between the randomised treatment groups, and in particular to investigate the effect of age on pharmacokinetics.

Background

The corticosteroid dexamethasone is a key component of childhood ALL therapy, with a favourable antileukaemic benefit having recently been reported over prednisolone induction therapy. However, while the use of dexamethasone has undoubtedly led to improvements in outcome seen over the past 10 years, it also makes a major contribution to a variety of short and long-term side effects which may negate its antileukaemic benefit. The toxicity observed in UKALL 2003 is seen as being unacceptably high in the context of a trial with such high disease free survival, with a >3% risk of treatment related mortality reported. In addition, approximately a quarter of patients suffer at least one non-haematological serious adverse event. Based on findings from this and previously published studies, the current study provides an opportunity to investigate the potential impact of pharmacokinetic variation in drug scheduling, i.e. standard versus short dexamethasone, on clinical outcome, including response and serious adverse events. Despite its successful use in the treatment of several haematological malignancies and other tumour types, very limited information is available concerning dexamethasone pharmacokinetics in children. A recently published study in this area showed substantial interpatient variability following treatment of children with ALL, with a greater than 10-fold variability in systemic drug exposure observed at a dose of 8mg/m²/day (Yang et al., 2008). Variability was correlated with a number of covariates including serum albumin concentration and concurrent use of other drugs, including doxorubicin and ketoconazole. In addition, the oral clearance of dexamethasone was greater in younger as compared to older children. Thus the older children experienced higher plasma concentrations of dexamethasone, consistent with an increased occurrence of toxicity in this age group. The influence of these key parameters on dexamethasone pharmacokinetics will be further investigated in the current study.

Patients and treatment

Blood samples for analysis of dexamethasone pharmacokinetics will be obtained from a total of 250 patients, male and female, receiving dexamethasone induction therapy in R1 as follows:

Group 1:NCI Standard Risk receiving Standard Dexamethasone (Regimen A)	(n=50)
Group 2:NCI Standard Risk receiving Short Dexamethasone (Regimen A)	(n=50)
Group 3:NCI High Risk receiving Standard Dexamethasone (Regimen B)	(n=50)
Group 4:NCI High Risk receiving Short Dexamethasone (Regimen B)	(n=100)*

Standard Dexamethasone: 6mg/m² for 28 days

Short Dexamethasone: 10mg/m² for a total of 14 days

* Group 4 will include patients receiving dexamethasone for 14 consecutive days (aged <10 years) in addition to those receiving dexamethasone on days 1-7 followed by days 15-21 (aged ≥10 years).

A minimum of 50 patients under the age of 5 years will be studied (≥25 receiving standard dexamethasone and ≥25 receiving short dexamethasone) to allow for potential differences in pharmacokinetics in younger patients to be investigated within the data analysis.

Pharmacokinetic sampling will be carried out on days 1, 2 or 3 and day 26, 27 or 28 (standard dexamethasone), day 1, 2 or 3 and day 12, 13 or 14 (short dexamethasone administered for 14 consecutive days) or day 1, 2 or 3 and day 19, 20, 21 (short dexamethasone administered on days 1-7 followed by 15-21) as described below. The actual dose administered to the patient and time of administration should be clearly recorded on the sampling sheet (see below) and it should be noted if this deviates in any way from the dose defined in the study protocol.

Sample Requirements

All patients must have a central venous catheter (single or multi-lumen catheter or portocath) or peripheral cannula in place in order for samples to be taken for pharmacokinetic analysis. Wherever possible, pharmacokinetic samples should be taken when clinical blood samples are obtained.

Blood samples (3ml) should be obtained pre-treatment and at 1, 2, 4 and 8 hours after the first dose of dexamethasone on days 1, 2 or 3 and day 26, 27 or 28 (standard dexamethasone), day 1, 2 or 3 and day 12, 13 or 14 (short dexamethasone administered for 14 consecutive days) or day 1, 2 or 3 and day 19, 20, 21 (short dexamethasone administered on days 1-7 followed by 15-21). Exact sampling times should be clearly recorded on the 'Dexamethasone PK Study Sampling Sheet' contained in the Investigator Site File.

A saliva sample will be collected using an age-appropriate kit supplied by Oragene® DNA (OG-500 & OG 575). These kits should be requested in advance from the Northern Institute for Cancer Research (NICR; please contact Gareth Veal/Julie Errington, Tel. 0191 208 4332 or 0191 208 4357). Samples should be collected according to the manufacturer's instructions and stored at room temperature or frozen at -20°C. DNA from these samples will be isolated and investigated for genetic variation in genes relevant to the pharmacology of dexamethasone, using techniques established in the NICR.

Blood samples should be immediately transferred to heparinised tubes, centrifuged for 5 min at 2,000 rpm and 4°C and the plasma obtained transferred to a clean labelled tube and stored at -20°C prior to transport to the Northern Institute for Cancer Research (NICR), Newcastle University.

Sample transport

All samples should be sent to the Northern Institute for Cancer Research (NICR) in a single package by overnight courier (Monday – Thursday), packed on dry ice in an insulated container, following completion of all pharmacokinetic and pharmacogenetic sampling.

The NICR should be contacted prior to the transport of samples to obtain details of the courier and reference number, and should also be notified on the day that the samples are sent (please contact Gareth Veal/Julie Errington, Tel. 0191 2086 4332 or 0191 208 4357).

Expenses will be paid by the NICR to cover all sample transport costs.

Assay

A fully-validated liquid chromatography/mass spectrometry (LC/MS) method for the measurement of dexamethasone in plasma is established in our laboratory in the NICR. The assay has a limit of detection of 0.05ng/ml and a lower limit of quantitation of 1.0ng/ml, allowing the reliable measurement of dexamethasone in plasma samples obtained from patients receiving the drug clinically.

Beclamethasone is used as an internal standard and dexamethasone calibration curves are linear between 1 and 2,000ng/ml, with correlation coefficients ≥ 0.996 . Intra- and inter-assay accuracy and precision data demonstrated percentage deviation from the theoretical mean (%DMT values) of $<10\%$ and $<6\%$ respectively ($n=10$). Samples obtained for pharmacogenetic analysis will be genotyped for the known functional polymorphisms in genes relevant to the pharmacology of dexamethasone. Appropriate techniques for these analyses are established in the NICR.

Statistical analysis

Statistical advice was provided by Michael Cole in the NICR. Power calculations are based upon a two group comparison of dexamethasone clearance, i.e. comparing standard versus short dexamethasone treatment. Clearance is assumed to be log-normally distributed with an SD of approximately 0.7. This SD value is taken from Yang et al (2008), who estimated inter- and intra-patient CV to be 0.46 and 0.53 respectively. Hence, total CV is approximately $\sqrt{0.46^2 + 0.53^2} = 0.70$. With a study population of 250 patient, the study would have $>90\%$ power to detect a 40% relative difference in dexamethasone clearance between the defined groups. Inclusion of a minimum of 50 younger children <5 years of age provides a 90% power to detect a 57% relative difference between younger and older patient cohorts.

Pharmacokinetic analysis

The data obtained will be used to determine pharmacokinetic parameters including area under the plasma concentration-time curve (AUC), clearance and half-life ($t_{1/2}$) for dexamethasone in each of the defined treatment groups. Dexamethasone pharmacokinetics in each arm of the randomisation and each dose regimen will be compared accordingly. Pharmacokinetic modelling will be carried out using these data in conjunction with patient characteristics and clinical parameters in order to investigate the key factors involved in determining individual drug exposures within the defined patient populations. The influence of pharmacokinetic parameters and genotype on day 2 MRD status, event free survival and grade III/IV toxicity will be investigated using Cox proportional hazards models and t-test analysis respectively.

References

1. Yang L, Panetta JC, Cai X, et al. Asparaginase may influence dexamethasone pharmacokinetics in Acute Lymphoblastic Leukaemia. J Clin Oncol 2008;26:1932-9.

APPENDIX 16 – INVESTIGATIONAL MEDICINAL PRODUCT (IMP) SUPPLIES

See protocol section 7.3 for a summary of the Investigational Medicinal Products (IMPs) used in this trial. Please refer to the UKALL 2011 Pharmacy Manual regarding IMP management and accountability recording.

Commercial supplies of licensed drugs taken from routine pharmacy stock are used for IMP doses. Any EU licensed brand may be used. Drugs used in this trial are not provided by the Sponsor and should be purchased through usual hospital purchasing arrangements.

NB: Unlicensed 'Special' formulations may not be used for IMP doses in clinical trials.

APPENDIX 17 – OSTEONECROSIS RECOMMENDATIONS

Background

The aetiology of osteonecrosis (ON) during ALL treatment is multifactorial. Dexamethasone plasma levels increase with age and can be increased by asparaginase-induced hypoalbuminaemia. Genetic factors are likely to play a part. Mechanisms of damage include direct osteocyte toxicity and embolic disease (hyperlipidaemia and altered coagulation).

Prospective MRI screening of hips and knees during ALL treatment, regardless of symptoms, has demonstrated that radiological evidence of ON is much commoner than previously thought. Relling et al (Ref 1) screened 365 patients in the Total XV protocol and have demonstrated a cumulative ON incidence of 71.8% (53.9% asymptomatic and 17.6% symptomatic). Only 26% of asymptomatic cases developed symptomatic ON by the completion of therapy. In the Total XV protocol (2000-2007) prednisolone is used during induction and no dexamethasone is given beyond day 100 during continuation/maintenance therapy. A small pilot study in Birmingham in 15 patients during the ALL2003 protocol has demonstrated a similar incidence of asymptomatic ON. Widespread ON affecting multiple areas was found even in the asymptomatic cases. Other important findings in the Relling study include an association between the development of ON and post-induction hypoalbuminaemia and hypercholesterolaemia, age >10years and intensive protocol arm.

Recommendations

1. Routine screening of asymptomatic cases is unnecessary in the absence of a prospective trial.
2. Asymptomatic ON detected coincidentally.
 - a) No evidence to suggest discontinuation of dexamethasone is routinely indicated in asymptomatic cases.
 - b) Monitor closely and early repeat MRI if symptomatic
 - c) Consider orthopaedic referral. The risk of collapse of the femoral head is affected by the location and extent of the necrotic lesion. All femoral head lesions which are either large or extend to the edge of the epiphysis should be referred to orthopaedic team for consideration of core decompression in order to prevent femoral head collapse. Using MRI images in both coronal and sagittal planes the Kerboul combined necrotic angle is a good MRI-based method to assess risk of hip collapse (Ref 2).
3. Symptomatic ON.
 - a) Consider continuation of dexamethasone and 6 monthly MRI screening to detect progression of ON.
 - b) For persistent/worsening symptoms or MRI progression, reduction/discontinuation of dexamethasone will need to be considered. If in doubt contact trial coordinators in these cases.
 - c) Consider orthopaedic referral (see 2c above)
 - d) Routine use of bisphosphonates can ONLY be recommended in patients with coexisting osteoporosis, defined by reduced bone mineral density and presence of low-impact fractures (ISCD Criteria) or as part of a clinical trial.

References

1. Pharmacokinetic, pharmacodynamic and pharmacogenetic determinants of osteonecrosis in children with acute lymphoblastic leukemia
Jitesh D. Kawedia, Sue C. Kaste, Deqing Pei, John C. Panetta, Xiangjun Cai, Cheng Cheng, Geoffrey Neale, Scott C. Howard, William E. Evans, Ching-Hon Pui and Mary V. Relling Blood First Edition Paper, online December 10, 2010; DOI 10.1182/blood-2010-10-31196
2. Prediction of collapse in femoral head osteonecrosis: a modified Kerboul method with use of Magnetic Resonance Images. Y.-C. Ha, H. J. Kim, S.-Y. Kim, K.-C. Kim, Y.-K. Lee, and K.-H. Koo. The Journal of Bone and Joint Surgery (American). 2006;88:35-40.

APPENDIX 18 – DIAGNOSIS AND MANAGEMENT OF PANCREATITIS

Acute pancreatitis is a well-recognized complication of asparaginase treatment for ALL. The incidence is between 5-10 %. Adolescents are up to four times more likely to develop asparaginase induced acute pancreatitis than younger children. Timing of onset of acute pancreatitis following asparaginase is variable.

Definition of pancreatitis in ALL 2011 (UK clinical guidelines; - Gut 2005, 54)

For the purpose of this trial pancreatitis is defined on the basis of at least two of the following features,

1. Abdominal pain strongly suggestive of acute pancreatitis
2. Serum amylase and/or lipase ≥ 3 times the upper limit of normal (lipase is preferred over amylase due to greater specificity)
3. Characteristic imaging findings of acute pancreatitis (ultrasonography is often unhelpful but contrast enhanced CT is useful for both confirming the diagnosis, determining severity, assessing complications, and for guiding potential percutaneous interventions).

Management

1. Analgesia and fluid resuscitation should be given
2. Avoid further asparaginase, as there is a high likelihood of recurrence.
3. Use of scoring systems (APACHE II, CT severity index) can help determine the likelihood of complications and the potential need for transfer to specialist units/ITU.
4. Potential long term complications include the development of diabetes mellitus, chronic pancreatitis, and pseudocysts and referral to a gastroenterologist is suggested

APPENDIX 19 – USE OF CRISANTASPASE (ERWINIA L-ASPARAGINASE) IN PATIENTS WITH SYSTEMIC REACTIONS TO PEGASPARGASE (ONCASPAR)

1. A licensed preparation of crisantaspase is now available, thus providing an effective alternative for patients with hypersensitivity to E.Coli asparaginase.
2. Crisantaspase should be used in place of Pegylated E. Coli asparaginase in the following circumstances:
 - Systemic hypersensitivity reactions to pegylated E.Coli asparaginase (pegaspargase (Oncaspar)). This includes patients with generalised rash with or without anaphylactic symptoms, but not those with only local pain or redness at the site of injection.
 - Patients with previously documented systemic reactions to E.Coli asparaginase should receive crisantaspase in any remaining asparaginase containing courses.
3. Each dose of pegaspargase (Oncaspar) should be replaced with 6 doses of 20,000 Units/m² crisantaspase given on Mondays, Wednesdays and Fridays.
4. Crisantaspase should be administered by intra-muscular injection. For older patients requiring large volumes, the individual dose may be split between two injection sites.
5. Please notify the trials office of patients switching to crisantaspase by indicating this on the relevant CRF for the treatment phase.

A table for converting from pegaspargase to crisantaspase can be found on the next page.

Table for converting from pegaspargase to crisantaspase

Pegaspargase	Crisantaspase
Induction (all regimens) Pegaspargase 1000 units/m ² intramuscular on day 4 and 18	Induction (all regimens) Day 4 N/A Day 18: crisantaspase 20,000 units/m ² intramuscular on day 18 and then Mon/Wed/ Friday for 6 doses in total
Consolidation Regimen C Pegaspargase 1000 units/m ² intramuscular on day 16 and 44	Consolidation Regimen C Crisantaspase 20,000 units/m ² intramuscular on day 16 and then Mon/Wed/ Friday for 6 doses in total Crisantaspase 20,000 units/m ² intramuscular on day 44 and then Mon/Wed/ Friday for 6 doses in total
Capizzi maintenance (Regimen C only) Pegaspargase 1000 units/m ² intramuscular on day 3 and 23	Capizzi maintenance (Regimen C only) Crisantaspase 20,000 units/m ² intramuscular on day 3 and then Mon/Wed/ Friday for 6 doses in total crisantaspase 20,000 units/m ² intramuscular on day 23 and then Mon/Wed/ Friday for 6 doses in total
Protocol M-A (Regimen C only) Peg Asparaginase 1000 units/m ² intramuscular on day 3 and 23	Protocol M-A (Regimen C only) Crisantaspase 20,000 units/m ² intramuscular on day 3 and then Mon/Wed/ Friday for 6 doses in total Crisantaspase 20,000 units/m ² intramuscular on day 23 and then Mon/Wed/ Friday for 6 doses in total
Delayed intensification (all regimens) Pegaspargase 1000 units/m ² intramuscular on day 4	Delayed intensification (all regimens) Crisantaspase 20,000 units/m ² intramuscular on day 4 and then Mon/Wed/ Friday for 6 doses in total
(Regimen C only) Pegaspargase 1000 units/m ² intramuscular on day 43	Crisantaspase 20,000 units/m ² intramuscular on day 43 and then Mon/Wed/ Friday for 6 doses in total

APPENDIX 20 – GENOMICS SUB-STUDY

Background

Cytogenetic analysis is an essential diagnostic test in childhood ALL where specific chromosomal abnormalities are used for treatment stratification (e.g. *iAMP21*, *t(9;22)/BCR-ABL1*) and others are known prognostic factors. Cytogenetics, FISH (fluorescence in situ hybridization) and RT-PCR currently provide the gold standard techniques for their detection within the routine diagnostic setting. More recently, studies have detected a large number of genetic abnormalities which are involved in the pathogenesis of the disease. These include *CRLF2* deregulation, *IGH@* translocations, deletions (aka micro-deletions or copy number alterations) of genes involved in B cell differentiation (e.g. *PAX5*, *IKZF1* etc) and cell cycle control (e.g. *CDKN2A/B*, *RB1*, etc) and somatic mutations in genes leading to constitutive activation of RAS signalling, impairment of B cell development, disruption of P53/RB1 signalling and JAK family members. In addition, gene expression profiling (GEP) has identified a novel subgroup (BCR-ABL-like) which is associated with a poor outcome. Although it is genetically heterogeneous recent studies suggest that at least some of these patients may be sensitive to the tyrosine kinase inhibitors currently used to treat BCR-ABL positive patients. The clinical and prognostic relevance of these new abnormalities has yet to be fully established. Moreover the majority of aberrations are not routinely screened for by regional genetic laboratories, as their accurate detection requires the use of novel high-throughput technologies.

Leukaemia Research Cytogenetics Group (LRCG)

Since 1992 the LRCG has been responsible for coordinating and overseeing cytogenetic analysis for paediatric ALL trials (UKALLXI, ALL97 and ALL2003) [Harrison et al (2001) *BJH* 113:3]. The LRCG has two functions. Firstly, to collect, review and classify cytogenetic results. This enables the investigation of the epidemiology and prognostic relevance of genetic abnormalities in ALL. Secondly, to use state-of-the-art technologies to discover and characterise new genetic abnormalities involved in the pathogenesis of ALL. In collaboration with the trial coordinators the LRCG has published numerous studies which have significantly advanced the field: e.g. Harrison et al (2005) *BJH* 129:520; Moorman et al (2007) *Blood* 109:2327; Moorman et al (2010) *Lancet Oncology* 11:429; Ensor et al (2011) *Blood* 117:2129.

Proposal for UKALL2011

The following strategy for organising genetic analysis in UKALL2011 has been agreed with the trial coordinators and the NCRI Childhood Cancer and Leukaemia Group (CCLG) Leukaemia Sub-Group (LSG).

The LRCG will continue to collect and review cytogenetic results on all patients treated on UKALL2011 using the established infrastructure through the network of cytogenetic laboratories. In addition, the LRCG will prospectively screen all patients for deletions and mutations in the major leukaemogenic genes. For BCP-ALL patients, this will include (1) deletions of *IKZF1*, *PAX5*, *EBF1*, *BTG1*, *RB1* etc; (2) mutations of *JAK2*, *IKZF1*, *PAX5*, *NRAS*, *KRAS2*, *TP53*, etc; (3) rearrangements involving *CRLF2*, *IGH@*, etc. For T-ALL and T-NHL patients, this will include (1) deletions of *TAL1*, *PHF6*, *LMO2*, *NF1*, *CDKN2A* etc; (2) amplifications of *MYB*, *NUP214-ABL1*, etc; (3) rearrangements involving T-cell specific oncogenes e.g. *TLX1*, *TLX3*, *TAL1* etc; (4) mutations of *PHF6*, *NOTCH1*, etc.

Copy Number Alterations (CNA) (deletions and amplifications) will be detected using Multiplex Ligation-dependent Probe Amplification (MLPA) which is a rapid multiplex PCR-based technique developed by MRC Holland. It allows comparative quantification of multiple genomic sites and is the optimum method for detecting the CNAs mentioned above. In association with MRC Holland, we have developed MLPA kits for BCP-ALL and T-ALL. Somatic gene mutations will be detected using a high-throughput mutation screen. Other gene rearrangements such as *CRLF2* deregulation and translocation involving the *IGH@* locus will be detected using a combination of FISH, MLPA, RT-PCR and Q-PCR. In addition, we will investigate the incidence and clinical relevance of the genomic rearrangements which give rise to the BCR-ABL-like signature. As these abnormalities may make the leukaemia sensitive to tyrosine kinase inhibitors, this study will help us evaluate the potential role of introducing these drugs into future protocols.

This comprehensive and prospective genetic screen of a large paediatric ALL cohort treated on a single clinical trial will be a world first. Therefore, the resulting dataset will allow us to evaluate the prognostic and predictive value of multiple genetic markers in paediatric ALL. The prospective nature of the screening will allow us to deliver results to the trial coordinators in an unprecedentedly timely fashion.

Requirements

Data collection and FISH testing: Routine diagnostic cytogenetic and molecular cytogenetic analysis will be performed locally and must include standard G-banded analysis, as well as FISH tests for *ETV6-RUNX1*, *BCR-ABL* and *MLL*. These tests must be carried out as soon as possible after diagnosis as positive findings may exclude the patient from the trial or alter therapy. Copies of all genetic reports and left-over fixed cell suspension must be sent to the LRCG as soon as possible. The fixed cells will be used for screening for additional gene rearrangements.

Centralised genomic screening: The LRCG will undertake screening for newly described abnormalities and subgroups as described above. At this stage, this screening constitutes research so results will not be sent to treating clinicians. In order to perform this research the LRCG will need, from all patients:

1. Left over cell suspension, DNA and RNA for the local genetic laboratory (see section 5.1)
2. Viable cells (~5 million) or DNA from the Bloodwise Childhood Leukaemia Cell Bank (if the patient has consented to the biobank)

LRCG Contacts

Leukaemia Research Cytogenetics Group (LRCG)
Northern Institute for Cancer Research
Newcastle University
Level 5, Sir James Spence Institute,
Royal Victoria Infirmary
Queen Victoria Road
Newcastle upon Tyne
NE1 4LP ☎ 0191 282 1320 (Professor Christine Harrison) or 0191 282 1323 (Professor Anthony Moorman)
✉ lrcg-clinical@newcastle.ac.uk
📠 0191 282 1326

Publications

It is anticipated that this project will result in several publications. All publications will be planned and prepared in consultation with the trial coordinators. The invaluable contribution of patients and parents will be appropriately acknowledged in all publications. In addition, all the treating clinicians and associated healthcare professional will be acknowledged. Copies of all papers will be made freely available to all contributors. In the case of patients and parents this will be via their treating clinician.

APPENDIX 21 –TPMT AND 6MP

The Thiopurine study closed to recruitment on implementation of Protocol version 3.0.

CPA accredited laboratories for the TPMT genotype assay include City Hospital, Birmingham (www.cityassays.org.uk) and Guy's and St Thomas' Hospitals (www.gsts.com).

TPMT genotype

The thiopurine drug mercaptopurine (6-MP) is central to successful therapy of lymphoid malignancy in children. The cytotoxic effect of 6-MP is exerted by drug-derived thioguanine nucleotides (TGNs). The rate of TGN formation from 6-MP is regulated by drug methylation. Methylation leads to formation of non-cytotoxic metabolites and so reduces the cytotoxic effect of 6-MP. Inter-individual variation in methylation is due to the inherited activity of the enzyme thiopurine methyltransferase (TPMT). TPMT activity is governed by genotype and possibly other more complex factors.

The clinical importance of the TPMT genetic polymorphism has been illustrated by many studies which have linked the inheritance of lower TPMT activities with thiopurine sensitivity, specifically bone marrow toxicity. This is particularly important in the 0.3% of UK patients homozygous for the low/low genotype i.e. patients who inherit two low activity variant alleles, who require marked reductions in 6-MP dosing. **These patients lack functional TPMT activity and should be identified as soon as possible after diagnosis through TPMT testing in a CPA approved laboratory. See section 7.12.2. NB It is imperative that TPMT status is derived from genotypic and NOT phenotypic analysis.**

Screening for TPMT*2 and the TPMT*3 family will detect over 95% of variant low activity alleles, the remaining low alleles being rare or novel variants. Measurement of genotype is possible in small blood samples and is not influenced by blood transfusions. The occurrence of TPMT deficiency is 1 in 300; 11% of patients will be heterozygotes, i.e carriers of a low allele, and have an intermediate TPMT activity whilst the remaining 89% are TPMT wild-type (high activity). The clinical impact of TPMT heterozygous or wild-type genotypes is less clear. Thiopurine studies carried out throughout ALL 97/99 and 2003 and notable findings include:

- (1) Measurement of TPMT activity at disease diagnosis will detect TPMT deficiency. Screening for the TPMT low activity alleles belonging to the TPMT*3 family and TPMT*2 will detect over 95% of variant alleles. In the light of the number of children who receive blood transfusions at disease diagnosis TPMT genotyping is the more robust determinant of TPMT status in this patient cohort.
- (2) The measurement of TPMT activity at disease diagnosis measures a decayed enzyme which reflects the anaemia of ALL. This activity does not reflect functional TPMT activity during chemotherapy. Other than the detection of absolute TPMT deficiency, care must be taken in interpreting any TPMT activity measurement obtained in an at-diagnosis sample. Therefore TPMT status must be assessed by genotype not phenotypic enzyme activity.
- (3) Severe, life-threatening bone marrow toxicity, due to the excess production of 6MP derived TGN metabolites, is precipitated by TPMT deficiency
- (4) Bone marrow toxicity can also develop in TPMT heterozygotes when taking standard doses of 6MP. From the analysis of ALL97/99 and ALL2003 data, about 50% of heterozygous children had difficulty tolerating 100% (75mg/sqm) 6MP and were managed on dosages ranging from 15 to 64 mg/sqm, median 38. However, the remaining 50% of heterozygotes could tolerate 100% 6MP. With the exception of the TPMT deficient child, all other children should follow the standard protocol directed 6MP dosage adjustments.
- (5) TPMT heterozygotes requiring dose reductions accumulated significantly higher TGNs concentrations than heterozygotes that could tolerate 100% 6MP.
- (6) Reduced 6MP doses can influence relapse-free survival. Within the ALL trials children taking 6MP who accumulated less than the trial median TGN level whilst taking less than 100% 6MP were at an increased risk of relapse ($p = 0.02$). This represents a cohort at reduced dosages in response to other toxicities and presents strong evidence that such dosage reductions should be guided by monitoring TGN levels.

- (7) Many children continue to have problems with tablet taking. The incidence of non-compliance with oral 6MP is about 10%, with 3% of children repeatedly having MP metabolite concentrations at, or below, the lower limit of detection.

Warning, TPMT activity at disease diagnosis. At disease diagnosis TPMT status should be assessed by TPMT genotype. The TPMT enzyme at diagnosis is decayed (see notable findings) and 50% of children fall in the heterozygous, intermediate activity, range as documented for non-ALL and control adults. The majority of these patients are not true heterozygotes; in this situation TPMT activity measurements give a false classification of TPMT status.

Interpretation of 6MP metabolite values

The major use of 6MP metabolite analysis is the investigation of compliance problems. CPA accredited laboratories for 6MP metabolite assays (ask for the thioguanine nucleotide, TGN assay) include City Hospital, Birmingham (www.cityassays.org.uk) and Guy's and St Thomas' Hospitals (www.gsts.com).

The 6MP metabolites

The 6-mercaptopurine (6MP) active cytotoxic metabolites are called thioguanine nucleotides (TGNs). In addition you may be given levels for methyl-mercaptopurine metabolites which are products of the TPMT (thiopurine methyltransferase) reaction.

For those of you with major problems with compliance/tablet taking, a complete absence (nil) of 6MP metabolites after prolonged periods of 100% 6MP gives a very clear message. For those of you faced with metabolite values the information below may help with the interpretation.

TGNs

The range of interpretative limits given from both City Hospital, Birmingham and Guys and St Thomas' is 235 to 450 pmol TGN/ 8×10^8 red cells; TGNs towards the upper limit (approximately 400 pmol TGNs) are the appropriate value for children with leukaemia. Many children will naturally accumulate over 400 pmol. If the blood cells counts are within protocol target values this is nothing to worry about. If cell counts are within the target range, only adjust 6MP dose based on protocol cell count guidelines, not on 6MP metabolite information.

Methyl-metabolites

The methyl-mercaptopurine nucleotide metabolites (abbreviated to MeMPN, MeMP or 6MMPN) are products of the TPMT reaction and can be used to help interpret the metabolite profile if compliance is questioned. Children with a wild-type TPMT genotype should accumulate high concentrations of these metabolites, and this may be the reason for low TGNs. The 6MMPNs have no direct cytotoxic action; but 6MMPNs can inhibit the production of the DNA base guanine so 6MP derived TGNs are effective at lower concentrations. Lower TGNs and high 6MMPNs re-assure you that the child is taking the tablets, follow the protocol directed 6MP dose adjustment guidelines.

Ignore the interpretative limit of >5,700 pmol 6MMPN given from both City Hospital, Birmingham and Guys and St Thomas'. High concentrations of 6MMPN metabolites are not unusual in children with ALL on 100% 6MP. In the UKALL trials the median value is 10,000 pmol, and the lower quartile value 5,000 pmol (i.e. 75% of children taking 100% 6MP have 6MMPN levels higher than 5,000 pmol).

APPENDIX 22 – T-CELL NHL EMERGENCIES

Management of the Large Mediastinal Mass and or Pleural Effusion

If there is **significant respiratory impairment**, e.g. orthopnoea or thoracic outlet compression, then, except for a blood count, all invasive diagnostic procedures including lumbar puncture should be postponed. If a large pleural effusion is present, it may be carefully relieved under local anaesthesia with a 16G Teflon needle and the fluid drained used for diagnosis.

Under no circumstances should a critically large mediastinal tumour with clinical symptoms of respiratory distress be treated surgically.

It is acceptable to start emergency cyto-reduction on the basis of radiological assessment in the absence of tissue diagnosis.

Emergency cytoreductive therapy should be combined with careful management of any consequent tumour lysis (see below). Regular anaesthetic and radiological assessments should be performed and diagnostic surgery scheduled in the window where it is safe anaesthetically but the tumour is not completely disappeared.

Emergency cytoreductive therapy

Emergency cytoreductive therapy with dexamethasone 6mg/m²/ day or Prednisolone 60 mg/m²/day should be used initially. In cases in which steroid therapy is not effective in a clinically appropriate time frame then patients should receive a single dose of cyclophosphamide 300mg/m². The use of this cytoreductive therapy will not exclude the patient from the trial as long as they are randomised with 3days of starting steroid (see 4.1.1).

Surgery should be postponed until clinical stabilisation is achieved. Regular anaesthetic and radiological assessments should be performed and diagnostic surgery scheduled in the window where it is safe anaesthetically but the tumour is not completely disappeared.

Although surgery should usually be as minimally invasive as possible it should be sufficient to gain enough material for comprehensive characterisation of the disease. In no case should the biopsy material be fixed completely, because the opportunity to completely characterise the tumour biologically would then be lost.

Prior liaison between the oncologist, pathologist and surgeon should be undertaken so that together they can decide what tumour specimens are required for histopathology/cytology

Tumour Lysis Syndrome

When lymphoma cells die, at least five major substances are released, which are eliminated only by the kidneys: purine metabolites (xanthine, hypoxanthine), uric acid, potassium and phosphate. If the product exceeds its solubility, then xanthine, hypoxanthine and uric acid can crystallise out. This takes place in the kidney tubules and collecting tubules. Phosphate can precipitate with calcium as calcium-phosphate in both the kidney tubules and in the tissue. The results are oliguria/anuria, tissue necrosis and hypocalcemia. The solubility of xanthine and uric acid is much higher in an alkaline milieu than in an acidic one, but the precipitation of phosphate with calcium is favored in an alkaline milieu. Hypoxanthine can also crystallise at a pH > 7.5. Alkalisiation of the urine can, therefore, also favor the precipitation of cell lysis products. If the uric acid, potassium, phosphate and/or creatinine levels are already increased before the start of cytoreductive therapy, then measures for controlling these substances should be started first, before active cytoreductive therapy is begun. The start of cytoreductive therapy, however, should not be postponed much longer than 24 hours. The most important measure is the initiation and maintenance of a high urine output (100 - 250 ml/m²/hour). If this is working well, then metabolic imbalances which require intervention are rare. If, in spite of sufficient hydration and diuretics, it is not possible to initiate and maintain a satisfactory urine output, then early haemodialysis should be instituted. This situation is probably due to either direct infiltration of the kidneys, an obstruction of the urinary tract due to lymphomatous compression or an established

urate-phosphate or calcium-phosphate nephropathy, or a combination of these pathological conditions. Hyperkalaemia is the most frequent, immediately life-threatening complication of acute cell-lysis syndrome. If potassium levels rise above normal or, in the case of an existing hyperkalaemia, do not fall quickly after starting measures for preventing respectively therapy of acute cell-lysis syndrome, a life-threatening hyperkalaemia can evolve within a few hours.

Prevention of acute Cell-Lysis Syndrome

hydration: 3 000 – 5 000 ml/m²/day (2.5 or 5% dextrose with 0.45% sodium chloride IV infusion)

Maintain specific gravity of the urine ≤ 1.010 and rigorous fluid balance: output = input – insensible losses

Bodyweight measurement twice a day to monitor fluid retention.

For insufficient urine output: Furosemide 1 - 10mg/kg/day in divided doses.

initially, and ensure no extra potassium is in the hydration; slight hypokalemia is well tolerated

Rasburicase 0.2mg/kg/day i.v. over 30 minutes, minimum for 3-5 days, depending on tumour size or WBC.

Alkalisiation of urine is not necessary and might worsen renal impairment because it enhances the precipitation of calcium-phosphate in the renal tubules

Oliguria/Anuria

Definition: Urine excretion < 50 ml/m²/hour in spite of: Frusemide 10mg/kg/day i.v. and hydration 130 - 200 ml/m²/hour

The definition of < 5 ml/m²/hour is not useful in this situation: It is more useful in these circumstances to evaluate the urine output in relation to the input.

Diagnostic Investigations:	Ultrasound:	* Obstruction of the urinary tract * kidney infiltration
	Biochem:	* potassium * uric acid * phosphate * calcium
	Urine:	* uric acid crystals * calcium-phosphate crystals

Indications for haemodialysis:

- potassium > 7 mmol/l or > 6 mmol/l and increasing, in spite of increased hydration and diuretics
- phosphate > 10mg/100 ml (5 mmol/l) or product Ca x P > 6.4 mmol/l
- urine excretion: < 50 ml/m²/1 hour in spite of Frusemide 10mg/kg/day i.v. and fluid input 130 - 200 ml/m²/hour
- high-grade or complete urinary tract obstruction on both sides

APPENDIX 23 – DIAGNOSTIC FLOW CYTOMETRY IN UKALL 2011

Flow cytometric diagnosis of ALL should be made according to local practice within a CPA approved laboratory. Recommended MINIMUM diagnostic antibody panels for are specified below.

Surface Markers
CD19, CD10, CD22
CD2, CD7, CD3
CD13, CD33 , CD117
CD34, HLA-DR, CD45
Intracellular Markers
cytCD3
cytCD22, cyt CD79a, cytlgM
MPO TdT

Identification of Early Thymocyte Precursor (ETP) ALL

In 2009 Campana and colleagues at St Jude showed T-ALL derived from early thymocytes which retain stem cell features carried a very poor prognosis (5 year EFS 10%). ETP ALL as defined by Campana makes up 12% of T-ALL and has a characteristic immunophenotype.-(CD1a(-), CD8(-), CD5(weak) with stem-cell or myeloid markers. The St Jude data has not been confirmed in Dutch studies or preliminary analysis of ALL 2003. Prospective collection of detailed standardised immunophenotype in T-ALL in ALL 2011 will allow this issue to continue to be addressed. Thus in ALL 2011 it is mandatory that all T-ALL identified using the markers above are also screened for ETP ALL as follows.

Mandatory Markers in T-ALL
cCD3 MPO
CD4, CD5, CD8
CD1a, CD56

APPENDIX 24 – VINCRISTINE PHARMACOKINETIC AND PHARMACOGENOMIC STUDY - CLOSED

The Vincristine Pharmacokinetic study closed to recruitment on **31st March 2018**.

Aims:

1. To characterize the onset, severity, and natural history of vincristine-associated neuropathy in children undergoing therapy for ALL.
2. To compare the severity of vincristine associated neuropathy in children with ALL randomized to every 4 week vs. no vincristine therapy during maintenance therapy.
3. To optimise vincristine dosing in paediatric ALL patients.

Background

One of the most active anti-leukaemic drugs is vincristine. The dose-limiting toxicity of vincristine is a peripheral neuropathy (VIPN) characterized by progressive motor, sensory, and autonomic involvement. This can be painful, affect mobility and impair long term strength and dexterity. Little is known about the prevalence and impact of VIPN in paediatrics. One major reason for sparse data has been the lack of a specific and sensitive neuropathy tool.

- 1) We plan to use a variant of Total Neuropathy Score (TNS) tool called the Pediatric TNSr-Vincristine to capture data on VIPN in children. This will enable us to assess the validity and reliability of this tool and generate important data on the incidence, severity and natural history of VIPN in ALL. We will take advantage of the unique design of UKALL 2011, which will randomise patients between either continuing with vincristine pulses or not during maintenance therapy, to evaluate the toxicity of vincristine therapy in this setting.
- 2) Genome wide association studies have identified markers which predispose to chemotherapy induced neuropathy, suggesting that it may be possible to prospectively identify which individuals are at highest risk of developing this toxicity. Two such markers (enzymes *CYP3A4* and *3A5*) metabolize vincristine. *CYP3A5* high expressers break down vincristine faster than *CYP3A4* expressers. Preliminary data suggest that the incidence and severity of VIPN is related to inheritance of the *CYP3A* polymorphism. Thus we plan to assess the relationship between single nucleotide polymorphisms related to vincristine metabolism and the development of neurotoxicity.
- 3) Vincristine pharmacokinetics are characterised by large inter-patient variability in volume of distribution, total body clearance and half-life, with up to 40-fold variability in vincristine pharmacokinetic parameters reported. This large variability in exposure to vincristine may affect anti tumour responses. Thus we plan to assess the relationship between vincristine pharmacokinetics, pharmacogenomics, toxicity and efficacy (using an end of induction response to therapy i.e. MRD).

Design

A total of 100 patients will be recruited to the study aimed at evaluating and identifying predictors of VIPN in children with ALL.

Inclusion Criteria

- (1) Paediatric patients between the ages of 5 and 18 years inclusive (The *Paediatric TNSr-Vincristine* has not been validated outside these age ranges).
- (2) New diagnosis of ALL.
- (3) English-speaking subject and guardian (It will be difficult to arrange interpreters for all physiotherapy assessments).
- (4) Informed written consent by parent/guardian or patient.

Exclusion criteria

- (1) Diagnosis of relapsed or second cancer.
- (2) History of other developmental disorders (e.g. Down syndrome, other chromosomal disorders).
- (3) History of other neuromuscular disorders (such as traumatic brain injury, cerebral palsy).
- (4) Non-English-speaking subject or guardian.
- (5) Family or personal history of peripheral neuropathy (such as Charcot Marie Tooth Disorder).
- (6) History of upper or lower extremity amputation.

Neurotoxicity Evaluation:

Neuropathy and neuropathic pain will be assessed when subjects are receiving vincristine using the Paediatric TNSr-Vincristine. The Paediatric TNSr-Vincristine is a multidimensional instrument that will be used to measure peripheral neuropathy symptoms and signs (physical examination findings). Modified from the original TNS for use in children receiving vincristine, the instrument assesses proximal extension of numbness, tingling and neuropathic pain, vibration and temperature sensation, strength and tendon reflexes.

These functional measures were selected based on extensive validation in children with ALL and their sensitivity to VIPN. Assessments have successfully been carried out in over 70 children with ALL in multiple US institutions. To minimise inter-observer variation, neuropathy assessments will be carried out by a paediatric physiotherapist trained in performing chemotherapy-associated neuropathy assessments. All physiotherapists will attend a centralised training session consisting of watching a video and hands on practice. Competency will be assessed by a paediatric neurologist. This will also include yearly training updates. Each assessment should take 10-15 minutes to perform and can be done in the outpatient setting. Children will be assessed at diagnosis (when they will not have any signs of VIPN) and thus will serve as their own controls.

Data Collection

Clinical data to be collected at each assessment will include:

1. Cumulative vincristine dose (mg/m^2).
2. Number of vincristine doses withheld, i.e. the number of vincristine doses missed due to VIPN.
3. Number of vincristine doses reduced and percentage dose reduction
4. Concomitant medications (not including chemotherapy).
5. All grade 3 or 4 liver toxicities reported for the enrolled patients and corresponding dates.

There will be seven time-points for collection of data:

- 1) At the time of initial diagnosis.
- 2) During consolidation (weeks 6-8, following administration of the first 5 vincristine doses).
- 3) After the 5th week of delayed intensification and before the onset of maintenance.
- 4) During maintenance cycle 1 day 29 (1st year of treatment).
- 5) During maintenance cycle 3 day 29 (2nd year of treatment).
- 6) During maintenance cycle 5 day 29 (3rd year of treatment).
- 7) End of treatment for boys and girls.

The first time point will be completed shortly after diagnosis before the onset of VIPN. For the other time points there will be a 1 month period to collect data (data must be collected at the designated time point or within 1 month after that time point). If the evaluation day also includes intrathecal chemotherapy, the evaluation will be done prior to the procedure.

Sample Requirements

All patients must have a central venous catheter (single or multilumen catheter or portacath) or peripheral cannula in place for samples to be taken for pharmacokinetic analysis. Wherever possible, pharmacokinetic samples should be taken when clinical blood samples are obtained.

Blood samples (3mls) should be obtained during Induction at the following time-points:

- (1) Before vincristine is administered.
- (2) 5-10 minutes after 1st dose of vincristine is administered.
- (3) 30-60 minutes after 1st dose of vincristine is administered.
- (4) 2-4 hours after 1st dose of vincristine is administered.
- (5) 24 hours after 1st dose of vincristine is administered.
- (6) 96 hours after 1st dose of vincristine is administered.

Thus up to a total of six samples will be collected per patient. Blood samples will be collected in heparinised tubes, centrifuged for 5 min at 2,000 rpm and 4°C and the plasma obtained stored at -20°C prior to transport to the Northern Institute of Cancer Research (NICR), Newcastle University (see below). Exact sampling times should be clearly recorded on the 'Vincristine Pharmacokinetic Study Sampling sheet' contained in the investigator Site File.

A saliva sample will be collected using age appropriate kit supplied by Oragene®DNA (OG-500 & OG 575 kits). These kits should be requested in advance from the Northern Institute of Cancer Research (NICR; please contact Gareth Veal/Julie Errington, Tel 0191 208 4332 or 0191 208 4357). Samples should be collected according to manufacturer's instructions and stored at room temperature or frozen at -20°C. DNA from these samples will be isolated and investigated for genetic variation in genes relevant to the pharmacology of vincristine, using techniques established at the NICR.

Sample transport

All samples should be sent to the Northern Institute for Cancer Research (NICR) in a single package courier (Monday- Thursday), packed on dry ice in an insulated container, following completion of all pharmacokinetic and pharmacogenomic sampling. The NICR should be contacted prior to the transport of samples to obtain details of the courier and reference number, and should also be notified on the day that the samples are sent (please contact Dr Gareth Veal or Julie Errington, Tel 0191 208 4332 or 0191 208 4357). Expenses will be paid by the NICR to cover all sample transport costs.

Sample analysis

A liquid chromatography/mass spectrometry (LC/MS) method for the measurement of vincristine in plasma has been developed in the NICR. Vincristine and M1, the major metabolite, are quantified in plasma by liquid-liquid extraction, LC/MS analysis and internal standardization (vinorelbine). The data obtained will be used to determine pharmacokinetic parameters including AUC, clearance and half-life ($t_{1/2}$) for vincristine and M1. Pharmacokinetic modelling will be carried out using these data in conjunction with patient characteristics and clinical parameters in order to investigate key factors involved in determining individual drug exposures within the defined patient populations. Samples obtained for pharmacogenetic analysis will be genotyped for the known functional polymorphisms in genes relevant to the pharmacology of vincristine. Appropriate techniques for these analyses are established in the NICR.

Sample Size and Power Calculations for Genetic Association Analyses

We have two primary hypotheses in this project. The first hypothesis is to test whether severity of vincristine associated neuropathy in children with ALL randomized to every 4 week vincristine is different from no vincristine therapy during maintenance therapy; the second hypothesis is to test whether CYP3A5 high expressers (*1/*1, *1/*3, *1/*6, *1/*7) and low expressers (*3/*3, *3/*7, *3/*6, *6/*7) experience different severities of neuropathy after vincristine treatment. The following table presents the power and effect size analysis (assuming a high CYP3A5 expresser frequency of ~15%):

Hypotheses	Sample sizes	Fold difference in TNS between two groups		
		1.5	2.0	2.5
Aim 1: maintenance vs. no-maintenance	50 vs. 50	0.40	0.88	0.98
Aim 2: CYP3A5 expresser vs. non-expresser	15 vs. 85	0.21	0.58	0.84

The type I error is controlled at 2.5% for each hypothesis. The sample size and power are calculated using a two-sided t-test.

Genotype/Phenotype Correlations

Primary Data Analysis:

Neuropathy scores measured with *The Paediatric* TNSr-Vincristine will be compared between the two maintenance arms (randomised to vincristine or not) using the t-test analysis. SNPs and their derived haplotypes will be correlated with vincristine pharmacokinetics (predominantly Clearance and AUC), neuropathy scores (maximum neuropathy score, month of onset of neuropathy and number of treatment months with neuropathy) and disease outcome (MRD-High/Low). Multivariate regression analyses will be performed. Specifically, genotype frequencies will be compared between patients who experienced grade 3/4 neurotoxicity versus patients who did not experience grade 3/4 neurotoxicity. Clinical, demographic and co-medications will be used as covariates in the regression analyses.

APPENDIX 25 – SCREENING FOR TARGETABLE TYROSINE KINASE ALTERATIONS IN UKALL2011 PATIENTS

The genetic characterisation of acute lymphoblastic leukaemia (ALL) has revealed a number of alterations which result in tyrosine kinase activation and sensitivity to a tyrosine kinase inhibitor (TKI).^{1,2} Typically these rearrangements involve the fusion of a kinase gene (e.g. ABL1, ABL2, CSF1R or PDGFRB) to wide variety of “activating genes” (e.g. PAX5, EBF1, NUP214, ZMIZ1, etc) and are referred to as “ABL-class fusions”. Although the majority of ABL-class fusions occur within B-cell precursor ALL, it is well known the NUP214-ABL1 amplification can occur in T-ALL³ and we have identified a case of T-ALL harbouring an ABL2 rearrangement. There is growing evidence that patients harbouring one of these fusions will respond to treatment with a tyrosine kinase inhibitor (e.g. imatinib or dasatinib).⁴⁻⁷

Preliminary analysis of ALL2003 data has revealed the following key information:

- Patients with high levels of MRD post induction (>1%) have a 6-fold and 3-fold increased risk of relapse compared to MRD negative and other MRD positive patients respectively.
- ~10% of B-cell precursor ALL patients with high levels of MRD after induction harbouring an ABL-class fusion.
- ABL1 and ABL2 rearrangements can occur in T-ALL

These findings correlate with unpublished data emerging from other clinical trial study groups, including the Children’s Oncology Group (USA), Dutch Children’s Oncology and the Austrian and Italian BFM study groups.

Therefore, in light of these findings we have decided to revise the screening algorithm for detecting these targetable abnormalities (see flow chart below). Those patients who should be screened for an ABL-class fusion should have **BOTH** of the following clinical and genetic criteria:

Clinical criteria:

- Induction failure
 - o Failure to achieve a complete remission
 - o M2 or M3 marrow at the end of induction
- MRD level of >1% after induction
- MRD level of >0.5% at week 14

Genetic criteria:

- B-other ALL*
- T-ALL

*Definition of B-other ALL - B-cell precursor ALL without one of the following:

- t(12;21)(p13;q22)/ETV6-RUNX1
- high hyperdiploidy (51-65 chromosomes)
- t(9;22)(q34;q11.2)/BCR-ABL1
- KMT2A (MLL) rearrangement
- t(1;19)(q23;p13)/TCF3-PBX1
- t(17;19)(q23;p13)/TCF3-HLF
- near-haploidy (<30 chromosomes)
- low hypodiploidy (30-39 chromosomes)
- intrachromosomal amplification of chromosome 21 (iAMP21).

Please note, as the screening is focussed on patients who are not responding well to therapy, it is the responsibility of the treating clinician to initiate the request for this screening.

Screening recommendations for regional genetic laboratories.

- PDGFRB/CSF1R

Rearrangements involving the PDGFRB and CSF1R genes, which are juxtaposed on the long arm of chromosome 5, can reliably be detected using a commercially available dual colour FISH break apart, for example the Vysis PDGFRB Break Apart FISH Probe Kit.

Therefore we recommend screening eligible B-other/T-ALL UKALL2011 cases with FISH probes for PDGFRB.

- ABL1

Virtually all reported rearrangements involving ABL1 result in a split ABL1 signal using a commercially available dual colour FISH probe such as the Vysis LSI BCR/ABL Dual Color Dual Fusion Translocation Probe Kit. The exceptions are cases of ETV6-ABL1 arising from insertions. However, a recent collaborative study has shown that the vast majority of ETV6-ABL1 cases display abnormal signals patterns with either a BCR-ABL1 or a ETV6-RUNX1 probe.⁸

Therefore we recommend screening all UKALL2011 cases with FISH probes for BCR-ABL1 and ETV6-RUNX1.

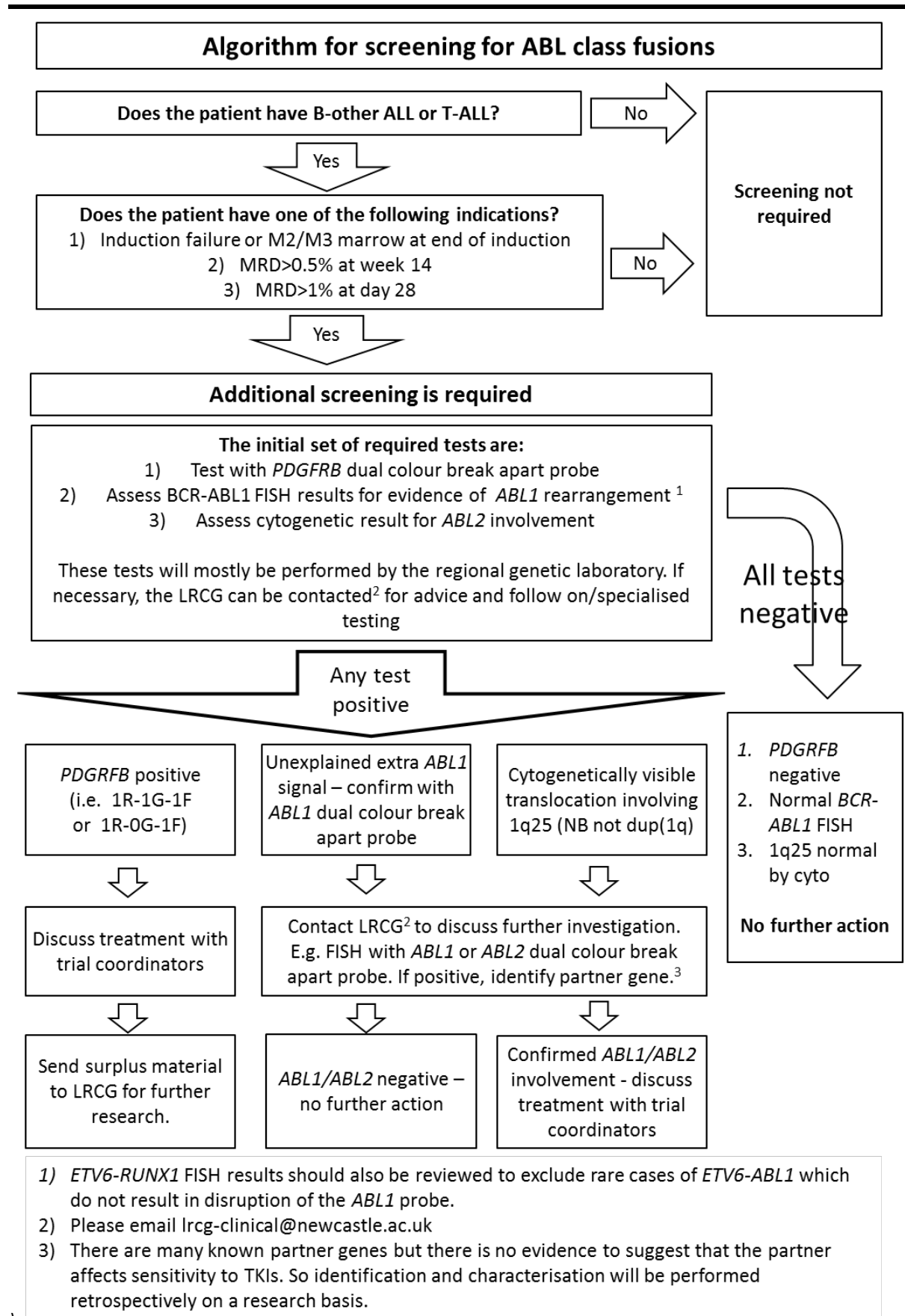
- ABL2

To date, very few ABL2 translocations have been reported; <5 cases worldwide. The LRCG has screened >360 B-other ALL cases and only one translocation has been identified. Moreover, cytogenetic analysis of this case showed a 1q25 breakpoint. One additional T-ALL case, which also had a 1q25 breakpoint by cytogenetics, has also been identified.

Therefore currently we recommend reviewing the cytogenetics of eligible B-other/T-ALL UKALL2011 cases for abnormalities on 1q25.

References

1. Roberts KG, Li Y, Payne-Turner D, et al. Targetable kinase-activating lesions in Ph-like acute lymphoblastic leukemia. *N Engl J Med*. 2014;371(11):1005-1015.
2. Roberts KG, Morin RD, Zhang J, et al. Genetic alterations activating kinase and cytokine receptor signaling in high-risk acute lymphoblastic leukemia. *Cancer Cell*. 2012;22(2):153-166.
3. Graux C, Stevens-Kroef M, Lafage M, et al. Heterogeneous patterns of amplification of the NUP214-ABL1 fusion gene in T-cell acute lymphoblastic leukemia. *Leukemia*. 2009;23(1):125-133.
4. Weston BW, Hayden MA, Roberts KG, et al. Tyrosine kinase inhibitor therapy induces remission in a patient with refractory EBF1-PDGFRB-positive acute lymphoblastic leukemia. *J Clin Oncol*. 2013;31(25):e413-416.
5. Lengline E, Beldjord K, Dombret H, Soulier J, Boissel N, Clappier E. Successful tyrosine kinase inhibitor therapy in a refractory B-cell precursor acute lymphoblastic leukemia with EBF1-PDGFRB fusion. *Haematologica*. 2013;98(11):e146-148.
6. Schwab C, Andrews R, Chilton L, et al. EBF1-PDGFRB Fusion in Paediatric Acute Lymphoblastic Leukaemia (ALL): Genetic Profile and Clinical Implications. *Blood*. 2014;124(21):1068.
7. Schwab C, Andrews R, Chilton L, et al. SSBP2-CSF1R Is a Recurrent Fusion in B-Other Acute Lymphoblastic Leukaemia with Variable Clinical Outcome. *Blood*. 2014;124(21):3773.
8. Zaliouva M, Moorman AV, Cazzaniga G, et al. Characterization of leukemias with ETV6-ABL1 fusion. *Haematologica*. 2016;submitted.



APPENDIX 26 – CHEMOTHERAPY –ASSOCIATED NEUROTOXICITY STUDY

Aims

The overall aim of this study is to identify biomarkers and therapeutic targets for adverse neurological outcomes in patients enrolled in UKALL 2011 (Registered & randomised patients)

To achieve this objective two cohorts of patients will be recruited:

- **Group 1** – A prospective cohort of children and young adults recruited at the time of original diagnosis of leukaemia/LBL and followed longitudinally for signs of developing subacute /chronic neurotoxicity
- **Group 2** - A cohort of children and young adults recruited during the course of their treatment who have developed significant acute neurotoxicity in the form of methotrexate associated stroke-like syndrome, posterior reversible encephalopathy syndrome and/or seizures (SPS)

Within these two groups, the following specific aims will be addressed:

- **Group 1 – Prospective cohort of children and young adults enrolled at diagnosis and followed longitudinally:**
 - o ***Specific aim 1:*** To use a software programme (CogState) to identify participants with early signs of neurocognitive toxicity
 - o ***Specific aim 2:*** To identify cerebrospinal fluid (CSF) biomarkers that predict adverse neurological outcomes.
 - o ***Specific aim 3:*** To identify whether polymorphisms in genes that control drug transport/metabolism or vulnerability of brain tissue to toxic damage can predict which patients are at high-risk of adverse neurological outcomes.
- **Group 2 – Cohort of children and young adults that fulfil the definition for SPS**
 - o ***Specific aim 1:*** To identify risk factors for SLS, the natural history of this disorder and predictors of incomplete recovery.
 - o ***Specific aim 2:*** To identify whether polymorphisms in genes that control drug transport/metabolism or vulnerability of brain tissue to toxic damage are associated with the development of SPS.

Background and rationale

Neurotoxicity during treatment for childhood ALL/LBL remains a significant problem. It can be acute as in the “stroke-like syndrome” seen with methotrexate^{1,2}, or may result in chronic neurocognitive defects³⁻⁵. Neurocognitive impairment is estimated to occur in between 20-50% of long-term survivors⁶. Although mean effects on global measures such as IQ are moderate, this hides a significant number of severely affected individuals³ and the risk factors for severe impairment are poorly understood. Recent studies have highlighted that neurocognitive impairment may continue to evolve throughout adulthood, even with chemotherapy-only regimes⁶. Worryingly, reported imaging and neurocognitive results⁴ are similar to those seen in early-onset dementia and the burden of neurological late effects may worsen over the next few decades. Thus, there is an urgent need to better understand the pathophysiology of neurotoxicity in childhood ALL, develop ways of identifying children at risk and to devise new treatments and/or protective measures.

For those without overt neurotoxicity symptoms, identification of patients at risk for poor neurocognitive outcomes might be achieved using early sensitive measures of neurocognition. Long-term follow up studies indicate that children treated for ALL/LBL have particular problems with memory, executive functions, attention and processing speed⁶. Whether these can be picked up early enough to act as a biomarker for long-term adverse outcome is unknown and will be tested in this study. Alternatively, biomarkers in cerebrospinal fluid may indicate high levels of toxic metabolites or biochemical markers of neurological damage that precede neurocognitive effects. The most highly implicated neurotoxic drug is methotrexate¹. Methotrexate pharmacokinetics do not correlate with neurotoxicity but secondary metabolites such as folate, homocysteine, adenosine and glutamate analogues have been implicated in both acute and chronic toxicity. An animal model of methotrexate

neurotoxicity⁷ suggests that excitotoxic glutamate analogues may cause neurotoxicity by binding to N-methyl D-aspartate (NMDA) receptors⁸. Importantly, in this pre-clinical model, NMDA antagonists can reverse and/or prevent neurotoxicity⁸ when administered concurrently with methotrexate⁹. This raises the possibility that CSF biomarkers will not only identify children at risk of neurotoxicity but also inform therapeutic strategies.

Finally, in the era of personalised medicine an alternative approach is to develop a panel of genetic predictors of neurotoxicity, which can prospectively identify high-risk children prior to any damage. Several candidate polymorphisms for chemotherapy-induced neurotoxicity have been proposed using small cohorts of patients with diverse cancer types. We will test these prospectively in a large standardised patient cohort. In addition, patients exhibiting overt chemotherapy related neurotoxicity such as those with SPS are likely to exhibit the highest hazard ratios for genetic predisposition and therefore their inclusion in this study will strengthen the likelihood of finding clinically relevant and actionable polymorphisms.

Overall, we predict that this study will provide significant insights into potential neurocognitive, biochemical and genetic biomarkers for neurotoxicity and provide strong underpinning science to determine whether NMDA antagonists are suitable for prevention and/or treatment of these potentially devastating neurotoxic effects

Study Design

Group 1 – Prospective cohort of children and young adults enrolled at diagnosis and followed longitudinally

Patients aged 4-25 years enrolled in UKALL 2011 will be eligible for this prospective longitudinal study of neurocognitive outcomes and their correlation with CSF biomarkers and genetic polymorphisms. Recruitment target is 150 patients with analysable data sets (at least 4 time points completed)

Recruitment will be from selected UKALL sites.

Inclusion Criteria:

- (1) Patients aged between 4-25 years inclusive at time of study consent (CogState is not validated for use in children aged <4years).
- (2) New diagnosis of ALL/LBL.
- (3) Participating in UKALL 2011
- (4) Informed written consent by patient or parent/guardian.
- (5) At least one parent able to speak English.

Exclusion Criteria:

- (1) Documented history of neurodevelopmental disorder prior to the diagnosis of ALL/LBL (e.g. Down syndrome, other chromosomal disorders).
- (2) Significant visual impairment preventing computer use.
- (3) Diagnosis of relapsed or second cancer.
- (4) Active meningitis or seizures less than one month from study enrolment
- (5) Patients whose Baseline line and Follow Up 1-4 [FU1-4] lumbar punctures will not be performed in the study centre

Group 2 – Children and young adults with overt chemotherapy related neurotoxicity – stroke-like syndrome, posterior reversible encephalopathy syndrome and/or seizures (SPS)

Recruitment will be from any UK site and the patient may be any age.

Inclusion Criteria:

- (1) Documented central neurological toxicity thought to be related to chemotherapy such as methotrexate stroke-like syndrome (SLS) Posterior reversible encephalopathy syndrome (PRES) or seizures with no clear alternative cause (see exclusion criteria below). Criteria for inclusion are summarised in table 1. Patients must fulfil both essential criteria, additional criteria are included to aid identification and classification of cases

- (2) Informed written consent by patient or parent/guardian
- (3) At least one parent able to speak English
- (4) Participating in UKALL 2011

Exclusion Criteria:

- (1) Patients with cerebral venous sinus thrombosis as a cause of their neurological symptoms
- (2) Patients whose symptoms are due to peripheral neuropathy or myopathy
- (3) Patients with clear cause for neurological event unrelated to chemotherapy neurotoxicity e.g. head injury following trauma, acute meningitis, viral encephalitis with known causative organism, seizures secondary to severe electrolyte imbalance or hypoglycaemia

Table 1: Definition of probable chemotherapy related neurotoxic event (SPS)

Essential criteria – must fulfil 1 and 2	
1.	New onset of one or more of the following neurological symptoms/signs: • Paresis • Aphasia/dysarthria • Altered mental status- somnolence, confusion, disorientation, emotional lability etc • Headache • Visual disturbances • Movement disorder • Loss of consciousness • Bilateral weakness • Seizures
2.	No other identifiable cause (such as migraine, cerebral venous sinus thrombosis, hypoglycaemia, electrolyte disturbance, acute meningitis/viral encephalitis)
Additional supportive criteria – Stroke-like syndrome	
Stroke-like syndrome patients should also fulfil all of the following criteria: A. New onset of one or more of the above neurological symptoms/signs within 21 days of MTX therapy (intrathecal or intravenous) B. EITHER: Findings of characteristic white matter changes of leukoencephalopathy on MRI (see below) AND/OR: Characteristic clinical course with waxing and waning symptoms usually leading to complete resolution over 1-7 days (Note in severe cases only partial resolution may be seen in this time frame) C. Isolated seizures without accompanying additional neurological features from the list above and without criteria A and B above are excluded) Characteristic MRI findings (best seen on T2-weighted images or ideally with diffusion weighted imaging (DWI)) include: oval shaped lesions of the subcortical white matter, often in the frontal, or parietal areas and often not conforming to a vascular territory. Lesions are generally hyperintense on DWI with a reduced apparent diffusion coefficient (ADC). <i>Note that imaging changes may be transient and therefore missed if the scan is delayed for a few days after onset of symptoms.</i>	
Additional supportive criteria – Posterior reversible encephalopathy syndrome	
PRES patients should also fulfil the following imaging criteria: Imaging findings: best imaging tool: contrast-enhanced MRI and DWI – Best diagnostic clue: patchy (and confluent) cortical (and subcortical) territory lesions – Most common location: cortex, subcortical white matter – Predilection: parietal and occipital lobes, cerebellum; less common: basal ganglia, brainstem, frontal lobes – Size: extent of abnormalities highly variable – Mass effect: minimal or none – Enhancement: minimal or none	

– MRI findings:

T1WI: hypointense cortical / subcortical lesions

T2WI: hyperintense cortical / subcortical lesions

On DWI images PRES is usually normal or hyperintense. ADC is usually increased

Additional supportive findings (non-essential):

1) Timing: usually during the first 3-4 months of therapy (mostly during remission induction therapy)

2) Presence of arterial hypertension

3) Complete resolution of clinical/imaging findings, although MR findings may persist in some patients

4) EEG: usually non-specific alterations, sometimes epileptiform discharges in the posterior region of the brain may be visible.

Study Protocol

1. Study time-points

Group 1 – Prospective cohort of children and young adults enrolled at diagnosis and followed longitudinally

Patients will be evaluated at 5 time-points [Baseline – Follow Up 1-4 [FU1-4] during therapy using a computer-based short age-appropriate neurocognitive test battery (CogState) and paired CSF samples taken at the time of routine scheduled lumbar punctures. In addition one saliva sample (or stored DNA from a remission bone marrow sample extracted during routine trial procedures) will be collected as a source of germline DNA and a clinical questionnaire will be completed at study entry and at completion of the study. Note patients randomised to Protocol-M will not have any further intrathecal therapy following Follow Up 2 (FU2) and therefore will only have CSF samples sent at Baseline – FU2 but should complete CogState testing at all time-points. See flowchart 1 for a summary of this test schedule.

1. **Baseline** – Baseline within 2-3 weeks of start of induction – clinical questionnaire, CogState test, CSF sample, saliva for DNA or remission BM for DNA.
2. **FU1** – During consolidation phase - CogState test, CSF sample.
3. **FU2** – End of interim maintenance/protocol-M - CogState test, CSF sample.
4. **FU3** – During first maintenance cycle - CogState test (all participants), CSF sample (omit if Protocol-M arm).
5. **FU4** – Approximately 2 years from start of therapy - CogState test and clinical questionnaire (all participants), CSF sample (omit if Protocol-M arm).

Further details of exact timings/test windows are given in the study guidelines.

To reduce participant burden and facilitate data and sample collection all assessments will be performed during routine scheduled hospital visits.

Note if patients in Group 1 experience a chemotherapy related neurological event (Stroke-like syndrome, PRES or seizures) they will switch to the group 2 study schedule described below

Group 2 – Children and young adults with Stroke-like syndrome/PRES and /or seizures(SPS)

Following a diagnosis of SPS, patients and their families can be approached for informed consent to enter this study. If consent is obtained the following will be collected:

1. SPS – Baseline – Ideally within 4 weeks of diagnosis of SPS - CogState test (once medically stable), clinical questionnaire, CSF sample (if LP performed), saliva (or remission BM) for DNA.
2. SPS FU1 – approximately 4 months after diagnosis of SPS - CogState test, clinical questionnaire, CSF sample (if LP scheduled) – note if CSF sample is sent CogState testing should be performed within 7 days of the CSF sample where possible.

3. SPS FU2 – approximately 1 year after diagnosis of SPS - CogState test, clinical questionnaire, CSF sample (if LP scheduled).
4. SPS FU3 onwards – testing should continue at 6 monthly intervals until completion of ALL treatment - CogState test, clinical questionnaire, CSF sample (if LP scheduled).

Note there is no lower age limit for group 2. Cogstate testing should only be performed if the child is 4 years or older at the time of testing

See flowchart 1 for a summary of this test schedule.

In addition, the results of any brain imaging performed as part of the routine clinical assessment of these patients will be collected.

2. Study procedures

CogState

Neurocognitive toxicity will be assessed using a state-of-the-art software programme – CogState - run on a standard laptop (with internet access). This programme is validated for children aged ≥ 4 years, shows good correlation with conventional neuropsychological tests, reliability and stability and is currently used in Dana Farber Cancer Institute (DFCI) 11-001 and Children's Oncology Group (COG) AALL1131 protocols. The tests target neurocognitive domains maximally affected by ALL/LBL treatment: processing speed, attention, visual recognition memory, working memory and executive ability⁶. Importantly, CogState tasks show negligible practice effects and so are suitable for this type of longitudinal study. The test battery varies with the age of the patient on the day of testing:

Age 4-5 years (inclusive) - 2 test battery - takes 5-10 minutes to complete.

Age 6-9 years (inclusive) - 5 test paediatric battery - takes 15-20 minutes to complete.

Age 10-25 years (inclusive) - 5 test standard battery - takes 15-20 minutes to complete.

Test administration is straightforward and can be supervised by any suitable hospital employee of any grade (nurse, doctor, play therapist, support assistants etc) as long as they have completed brief online training.

Clinical Questionnaire

Information will be collected on demographics (age, sex), any previous or existing neurological symptoms or cognitive problems and any family history of cognitive problems and results of any informative tests.

CSF biomarkers

2-3 mL of CSF (collected at the time of a therapeutic lumbar puncture, just prior to the administration of intrathecal chemotherapy) will be obtained at the designated time points. Samples should be immediately protected from the light placed on ice and then centrifuged (5 minutes at between 1200-1500 RPM) within 1-2 hours of collection. The supernatant should be transferred into a cryovial and immediately frozen at -80°C . Shipping: samples will be batched and shipped to Dr Halsey, University of Glasgow.

A panel of metabolites related to drug metabolism and markers of neural damage will be measured by HPLC and ELISA. The relationship with CogState scores will be investigated. In addition, any reported acute neurological toxicity in this cohort (seizures, stroke-like syndrome etc) will be identified with SAE reports and correlated with CSF biomarkers.

Host genetic variation

Constitutional DNA will be isolated from a saliva sample. The saliva sample should be collected using an age appropriate kit supplied by Oragene®DNA (OG-500& OG-575kits). Samples should be collected according to manufacturer's instructions and stored at room temperature prior to posting to Dr Halsey, University of Glasgow. Alternatively, remission BM DNA can be provided as a source of constitutional DNA if more convenient.

Following DNA extraction a candidate gene approach will be used to investigate host variation in genes controlling drug transport and metabolism or vulnerability of brain tissue to toxic damage. Genotypes of the patients with significant reductions in CogState scores (test group 1) or documented SPS (test group 2) will be compared to patients with no impairment (control group) and the Wellcome Trust case-control consortium (WTCCC) (5000+ cases). Previous studies indicate 20% of patients will have significant impairment so this will produce a ratio of 4 leukaemia controls for every case. DNA extractions will be performed using standard methodologies. Single-nucleotide polymorphisms (SNP) genotyping will be conducted using allele-specific PCR. Thymidylate synthase (TYMS) genotyping will be performed in-house using gel electrophoresis to determine the number of repeats.

3. Analysis

Group 1:

Analysis will identify patients with a significant decline in CogState scores between baseline and FU3. FU3 and FU4 will be compared to assess whether scores are stable over time. Results will be used to test; 1) whether early declines in neurocognitive scores predict persistent (FU4) defects, 2) whether CSF biomarkers correlate with neurocognitive test scores, and 3) whether host SNPs can be used to predict significant neurocognitive impairment.

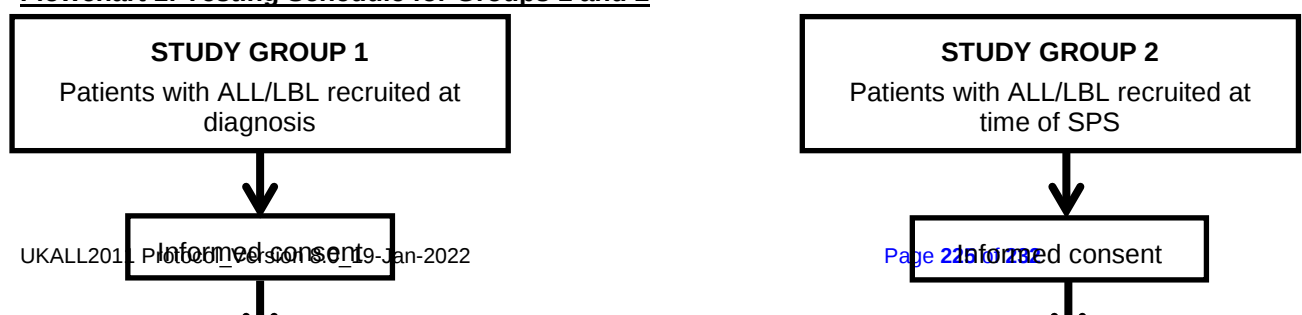
Group 2:

Analysis will define the initial CogState score after an SPS episode and then track the natural history of the impairment over time. In patients with CSF available, results will be used to test; 1) correlation between CSF biomarkers and neurocognitive test scores, 2) correlation between CSF biomarkers in patients with SPS and children from cohort 1 with no SPS. In patients with serial brain imaging available, results will be used to test; 1) correlation between CSF biomarkers and radiological severity, 2) correlation between serial CogState scores and radiological severity. Genomic analysis will address whether host SNPs can be used to predict children at risk of SPS (compared to unaffected controls from cohort 1).

Statistical considerations

Changes in individual patient Cogstate scores will be assessed using paired t-tests, a sample size of 100 has 80% power to detect an effect size of 0.28 ($p=0.05$). A Generalised estimating equation (GEE) approach will be used for repeated measurements at the 5 time-points and covariates will include treatment arm, risk-stratification, age, gender and ethnicity. Correlation between CogState scores and CSF biomarkers will be analysed using Pearson-product correlation analysis. Comparison with published studies indicates testing of ≥ 75 patients will have sufficient resolving power to detect important correlations between Cogstate results and CSF biomarkers. For genomic analysis; aiming for 40 cases with neurological impairment and 160 matched controls, we have $>90\%$ power to detect at $P<0.003125$ genetic effects with minor allele frequency >0.3 conferring allelic odds-ratios >3.0 , or $\sim 80\%$ power to detect genetic effects with minor allele frequency >0.2 conferring allelic odds-ratios >2.5 . (Power calculations carried out in QUANTO¹⁰, P value 0.003125 accounts for multiple testing (Bonferroni-correction) of 16 SNPs). Statistical analysis will be supervised by our collaborator Professor HJ Cordell.

Flowchart 1: Testing Schedule for Groups 1 and 2



References

1. Bond J, Hough R, Moppett J, Vora A, Mitchell C, Goulden N. 'Stroke-like syndrome' caused by intrathecal methotrexate in patients treated during the UKALL 2003 trial. *Leukemia*. 2013;27(4):954-956.
2. Rubnitz JE, Relling MV, Harrison PL, et al. Transient encephalopathy following high-dose methotrexate treatment in childhood acute lymphoblastic leukemia. *Leukemia*. 1998;12(8):1176-1181.
3. Halsey C, Buck G, Richards S, Vargha-Khadem F, Hill F, Gibson B. The impact of therapy for childhood acute lymphoblastic leukaemia on intelligence quotients; results of the risk-stratified randomized central nervous system treatment trial MRC UKALL XI. *J Hematol Oncol*. 2011;4:42.
4. Zeller B, Tamnes CK, Kanellopoulos A, et al. Reduced Neuroanatomic Volumes in Long-Term Survivors of Childhood Acute Lymphoblastic Leukemia. *Journal of Clinical Oncology*. 2013;31(17):2078-2085.
5. Waber DP, Queally JT, Catania L, et al. Neuropsychological outcomes of standard risk and high risk patients treated for acute lymphoblastic leukemia on Dana-Farber ALL consortium protocol 95-01 at 5 years post-diagnosis. *Pediatr Blood Cancer*. 2012;58(5):758-765.
6. Krull KR, Brinkman TM, Li C, et al. Neurocognitive Outcomes Decades After Treatment for Childhood Acute Lymphoblastic Leukemia: A Report From the St. Jude Lifetime Cohort Study. *J Clin Oncol*. 2013.
7. Li Y, Vijayanathan V, Gulinello M, Cole PD. Intrathecal methotrexate induces focal cognitive deficits and increases cerebrospinal fluid homocysteine. *Pharmacol Biochem Behav*. 2010;95(4):428-433.
8. Vijayanathan V, Gulinello M, Ali N, Cole PD. Persistent cognitive deficits, induced by intrathecal methotrexate, are associated with elevated CSF concentrations of excitotoxic glutamate analogs and can be reversed by an NMDA antagonist. *Behav Brain Res*. 2011;225(2):491-497.
9. Cole PD, Vijayanathan V, Ali NF, et al. Memantine protects rats treated with intrathecal methotrexate from developing spatial memory deficits. *Clin Cancer Res*. 2013.
10. Gauderman WJ. Sample size requirements for association studies of gene-gene interaction. *Am J Epidemiol*. 2002;155(5):478-484.

APPENDIX 27 TRECA (TRIALS ENGAGEMENT IN CHILDREN AND ADOLESCENTS) STUDY – OPEN AT SELECTED UKALL 2011 SITES

Aims

The overarching aims of TRECA are to evaluate the impact of multimedia information resources (MMIs) on:

- Trial recruitment and retention,
- Quality of decision-making about participation in healthcare trials involving children and adolescents.

Background and rationale

The effectiveness and safety of healthcare interventions is best determined through randomised controlled trials (RCTs) (1, 2). However, major barriers to the successful conduct and outcome of clinical trials are levels of recruitment and retention. In the UK, only a small proportion of trials actually recruit successfully to time and target (3). There is now international recognition of the importance of paediatric clinical trials to inform healthcare decisions for children and adolescents (4, 5). High quality trials involving children are essential to ensure that medication and treatments used in children are effective and safe (4, 6).

A potential barrier to recruitment and retention is the information provided to potential trial participants (2). How children and parents make decisions regarding participation in research and what information is important to them, both remain areas of uncertainty (7, 8). Conventionally, participant information about a trial is provided in printed form. However, these documents should be understandable to potential trial participants and assist their decision-making (9). However, the format of this information has received recurrent criticism, notably for being too long, difficult and technical (10-13). Furthermore, the content of trial sheets is mostly guided by regulatory agencies and can be inconsistent with what patients want to know (8, 10, 14). A number of studies report that trial participants do not understand information contained within participant information sheets (15) and that the information can be very wordy and overwhelming (10, 16). Potential participants who have lower levels of literacy are most likely to be affected by this (17). Furthermore, good graphic design is often lacking in participant information sheets, such as a structure that aids navigation of the information, and visual appeal to invite and engage the reader. For example, written information should inform a decision about participation, but may act more as a prospectus for the trial and as a contract between researchers and the participant (18). Re-writing, re-designing and user testing of trial information can produce an understandable and preferred resource (19).

An alternative for providing information to potential trial participants is through the use of an MMI (17, 20, 21), which is a website that presents key information using a combination of video, animation, text and diagrams. However, research is needed to identify and evaluate different ways of presenting multimedia information about research to children and parents (10). Multimedia presentation can be understood through reading, listening and watching and allows people with different preferences to use the resource effectively (17). The UKALL 2011 MMIs contain all key information that would be found in a written participant information about UKALL 2011 but will focus on information deemed important for children, adolescents and their parents (8, 20) when deciding whether to participate in a trial. Information will be available through animation, video clips, diagrams and written text.

Study design

Phase one of TRECA involved participatory design with children and adolescents with long-term health conditions, their parents and clinicians involved in paediatric trials. Interviews were undertaken to identify the needs and preferences for information about trials and the way they would like this information presented. This has informed the development of the MMIs. User testing was also undertaken to ensure that families could find information easily and understand the information.

Phase two of TRECA involves evaluating the MMIs in a series of embedded trials hosted within healthcare trials following the addition of host trial-specific content to the MMI. The MMIs will be tested for their impact on decisions about trial participation taken by children and adolescents and/or parents and behaviours (rates of recruitment to, and retention in, the host trials).

UKALL 2011 is one of the six trials to embed the TRECA MMIs. The evaluation phase will use an embedded randomised controlled trial design, with potential participants in the UKALL 2011 receiving one of three versions of the patient information.

UKALL 2011 will use a three-arm design (see Figure 1), in which individuals will be randomised to receive either:

- the standard written trial participant information sheet (PIS) alone,
- the standard PIS in addition to the MMI (or MMIs) or
- the MMI(s) only.

For those who receive both, TRECA will not determine the order in which participants access the PIS and MMI and will leave this for UKALL 2011 to determine, to suit the practical demands of patient recruitment.

In UKALL 2011, the randomisation of the TRECA arm will occur via sealed envelopes that are provided to each site by TRECA. Sites will be asked to document which arm of TRECA the participant received.

The UKALL 2011 MMIs will be presented by the recruiting clinician to those approached to participate in UKALL 2011. This will be done using a dedicated tablet computer provided to each participating UKALL 2011 site by TRECA. Recruiting clinicians will provide the MMI (or PIS, or PIS plus MMI) in the same way that they use the PIS during discussions. Participants will also be able to access the MMIs at home (via smartphone or a tablet or PC) and will be provided with the link to the relevant MMI.

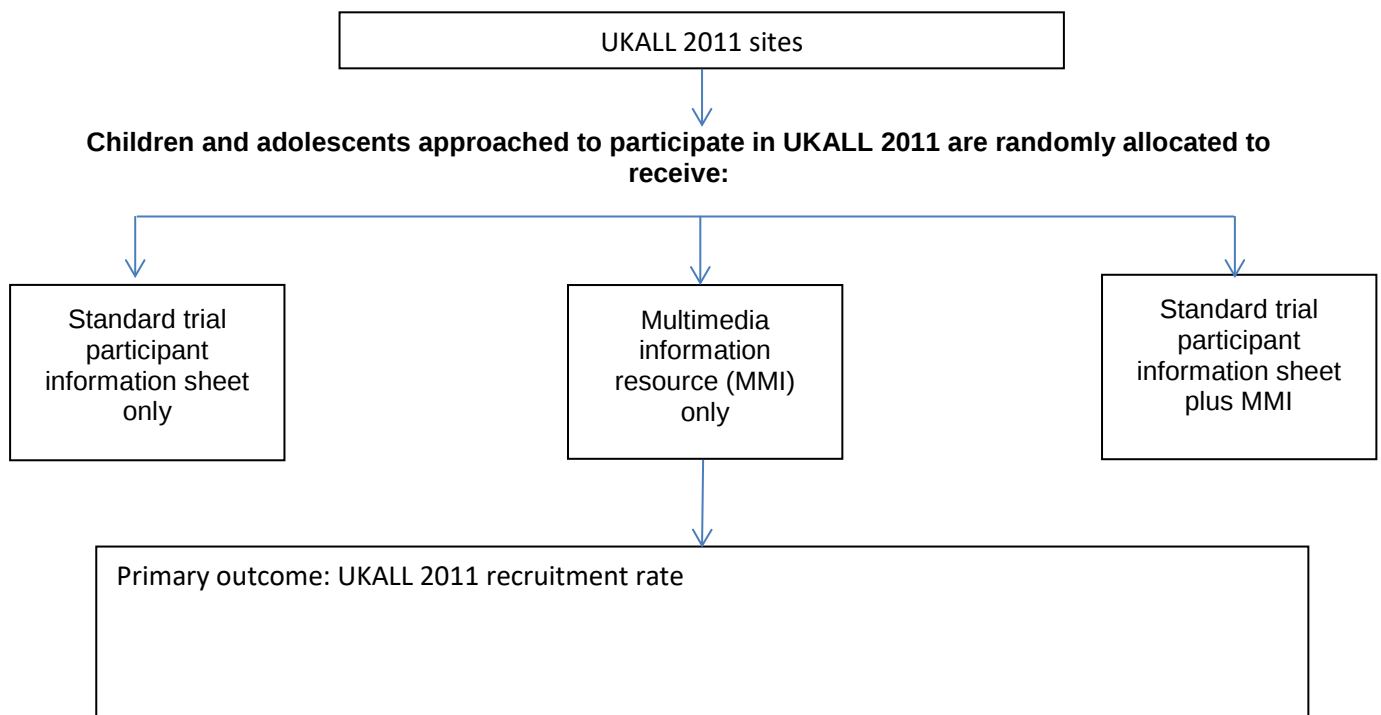


Figure 1: Summary of TRECA embedded within UKALL 2011

Inclusion criteria

- Patient must be eligible for R2 randomisation
- Family/patient were approached at Registration about involvement in TRECA
- Children and adolescents aged 8-18 years who have potential to contribute to a decision about consent (or assent) to trial participation
- Family has access to the internet (computer, tablet or smartphone)

Exclusion criteria

- Children and adolescents approached to participate in Randomisation 2 of UKALL 2011 who are either under the age of 8 years or older than 18 years of age
- Children and adolescents who are not able to participate in the decision-making about whether to take part in UKALL 2011
- Families who do not have access to the internet (computer, tablet or smartphone)

Outcome measures

Objective measures are the rates of recruitment to, and retention in, each host trial. We will also measure the quality of decision-making. The primary outcome will be recruitment rates.

For **recruitment**, we will calculate the proportion of patients who agree to participate from the total approached, for each arm of the embedded trial. We will assume that patient eligibility for host trial participation will have been assessed before an approach has been made. For the **retention** outcome we will obtain data on the number and timing of patient withdrawals from each host trial.

We will also measure the **quality of decision making** by potential host trial participants. Children and adolescents will be asked to complete a brief decisional scale, adapted from one used within the REFORM trial (22) and drawing conceptually on the SURE (23) and DelibeRATE scales. When a parent or parents have been involved in the participation decision, we will also ask them to complete the scale. As far as is possible, we will adapt the scale to facilitate completion by young children.

As far as is possible, we will aim to obtain decision quality scores both from individuals who decide to participate in the host trial and those who decline.

Data collection*Recruitment data*

Data on recruitment to the host trial will be recorded automatically within the host trial dataset. In order to assess any potential moderating influences of other variables on the effectiveness of the MMIs, we will aim to obtain data within each host trial of children and adolescents age, gender and postcodes (as a proxy for socio-economic status), according to allocation in the TRECA interventions and to UKALL 2011 trial participation decision.

Specifically the data requested by TRECA from trial sites will be anonymised data that includes for each patient:

- Screening ID and Participant ID (if patient consents)
- TRECA arm allocation (and order of PIS and MMI for those who receive both)
- Allocation within UKALL 2011
- Age (years) and gender of participant
- Age (years) and gender of parent providing consent (if applicable)
- Patient's postcode (or partial postcode or other measure for socio-economic status)
- Retention at delayed intensification stage of UKALL 2011

Decision-making questionnaires

For patients who decide to take part in UKALL 2011, the children and adolescents and/or parent will be asked to complete the decisional scale once the informed consent form has been completed. For

patients who decline participation in UKALL 2011, they will be asked to complete this measure in the clinic or will have them posted at home, as appropriate.

There are three decision-making questionnaires (with a participant being provided with, and completing, the appropriate version):

- Younger child version
- Older child/adolescent version
- Family version

Hard copy questionnaires will be provided to people approached to participate in UKALL 2011, as soon as is practicable after their decision about whether to participate in UKALL 2011 has been made. Questionnaires will be returned to the TRECA study office by post in self-addressed stamped envelopes by UKALL 2011 sites.

Analysis

The results from UKALL 2011 embedding the TRECA MMIs will be analysed. A joint UKALL 2011 and TRECA paper will be written based on these data and submitted to a peer-reviewed journal. It is anticipated that authors will comprise the UKALL 2011 TMG and TRECA Study Advisory Group members.

A meta-analysis of data from all six trials that embed TRECA will also be undertaken and submitted to a peer-reviewed journal to show overall impact of TRECA MMIs.

References

1. Treweek S, Lockhart P, Pitkethly M, Cook JA, Kjeldstrom M, Johansen M, et al. Methods to improve recruitment to randomised controlled trials: Cochrane systematic review and meta-analysis. *BMJ open*. 2013;3(2).
2. Ross S, Grant A, Counsell C, Gillespie W, Russell I, Prescott R. Barriers to participation in randomised controlled trials: a systematic review. *J Clin Epidemiol*. 1999;52(12):1143-56.
3. Bower P, Wilson S, Mathers N. Short report: how often do UK primary care trials face recruitment delays? *Fam Pract*. 2007;24(6):601-3.
4. Klassen TP, Hartling L, Craig JC, Offringa M. Children are not just small adults: the urgent need for high-quality trial evidence in children. *PLoS Med*. 2008;5(8):e172.
5. Joseph PD, Craig JC, Caldwell PH. Clinical trials in children. *Br J Clin Pharmacol*. 2013;79(3):357-69.
6. Britton A, McKee M, Black N, McPherson K, Sanderson C, Bain C. Threats to applicability of randomised trials: exclusions and selective participation. *J Health Serv Res Policy*. 1999;4(2):112-21.
7. Varma S, Jenkins T, Wendler D. How do children and parents make decisions about pediatric clinical research? *J Pediatr Hematol Oncol*. 2008;30(11):823-8.
8. Woolfall K, Shilling V, Hickey H, Smyth RL, Sowden E, Williamson PR, et al. Parents' agendas in paediatric clinical trial recruitment are different from researchers' and often remain unvoiced: a qualitative study. *PLoS ONE*. 2013;8(7):e67352.
9. Behrendt C, Golz T, Roesler C, Bertz H, Wunsch A. What do our patients understand about their trial participation? Assessing patients' understanding of their informed consent consultation about randomised clinical trials. *J Med Ethics*. 2011;37(2):74-80.
10. Caldwell PH, Dans L, de Vries MC, Newman Ba Hons J, Sammons H, Spriggs MBM, et al. Standard 1: consent and recruitment. *Pediatrics*. 2012;129 Suppl 3:S118-23.
11. Eder ML, Yamokoski AD, Wittmann PW, Kodish ED. Improving informed consent: suggestions from parents of children with leukemia. *Pediatrics*. 2007;119(4):e849-59.
12. Ogloff JR, Otto RK. Are research participants truly informed? Readability of informed consent forms used in research. *Ethics & behavior*. 1991;1(4):239-52.
13. Tarnowski KJ, Allen DM, Mayhall C, Kelly PA. Readability of pediatric biomedical research informed consent forms. *Pediatrics*. 1990;85(1):58-62.

-
14. Masty J, Fisher C. A Goodness-of-Fit Approach to Informed Consent for Pediatric Intervention Research. *Ethics & behavior*. 2008;18(2-3):139-60.
 15. Stead M, Eadie D, Gordon D, Angus K. "Hello, hello--it's English I speak!": a qualitative exploration of patients' understanding of the science of clinical trials. *J Med Ethics*. 2005;31(11):664-669.
 16. Shilling V, Williamson PR, Hickey H, Sowden E, Beresford MW, Smyth RL, et al. Communication about children's clinical trials as observed and experienced: qualitative study of parents and practitioners. *PLoS ONE*. 2011;6(7):e21604.
 17. Tait AR, Voepel-Lewis T. Digital multimedia: a new approach for informed consent? *JAMA*. 2015;313(5):463-4.
 18. Armstrong N, Dixon-Woods M, Thomas A, Rusk G, Tarrant C. Do informed consent documents for cancer trials do what they should? A study of manifest and latent functions. *Social Health Illn*. 2012;34(8):1230-45.
 19. Knapp P, Raynor D, Silcock J, Parkinson B. Performance-based readability testing of participant information for a Phase 3 IVF trial. *Trials*. 2009;10(1):79.
 20. Hutchison C, Cowan C, McMahon T, Paul J. A randomised controlled study of an audiovisual patient information intervention on informed consent and recruitment to cancer clinical trials. *Br J Cancer*. 2007;97(6):705-11.
 21. Shneerson C, Windle R, Cox K. Innovating information-delivery for potential clinical trials participants. What do patients want from multi-media resources? *Patient Educ Couns*. 2013;90(1):111-7.
 22. Knapp P, Graffy J, Bower P, Rick J, Fairhurst C, Rogers S, et al., editors. Preliminary analysis of a new measure of quality of patient decision making about research participation. Liverpool, UK, May 2017: Presented at the 4th International Clinical Trials Methodology Conference.
 23. Parayre AF, Labrecque M, Rousseau M, Turcotte S, Légaré F. Validation of SURE, a four-item clinical checklist for detecting decisional conflict in patients. *Med Decis Making*. 2013;34(1):54-62.