Imperial College Healthcare NHS Trust

Imperial College London

PREGRRS

Pregnancy Reference Range Study

V2.0

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FUNDERS:	N/A
STUDY COORDINATION CENTRE:	Imperial College Healthcare NHS Trust
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Protocol authorised by:

Name & Role	Date	Signature
Professor Tricia Tan (PI)	02/10/2020	
Dr Rebecca Scott (Co-I)	02/10/2020	
Dr Mariana Abdel-Malek (Co-I)	02/10/2020	
Dr Bryony Jones (Co-I)	10/01/2020	

Study Management Group

Chief Investigator:	Professor Tricia Tan (ICHNT & ICL)
Co-investigators:	Dr Rebecca Scott (ICHNT & ICL), Dr Bryony Jones (ICHNT)
Statistician:	Following National and International Guidance for production of reference ranges
Study Management:	Dr Rebecca Scott (ICHNT & ICL) Dr Mariana Abdel-Malek (ICHNT)

Study Coordination Centre

For general queries, supply of study documentation, and collection of data, please contact:

Study Coordinator: Dr Rebecca Scott
Address: Department of Diabetes & Metabolism
Hammersmith Hospital,
Du Cane Road, London, W12 0NN
Tel: 07970742784 E-mail: Rebecca.scott22@nhs.net

Clinical Queries

Clinical queries should be directed to Dr Rebecca Scott who will direct the query to the appropriate person

Sponsor

Imperial College Healthcare NHS Trust is the main research Sponsor for this study. For further information regarding the sponsorship conditions, please contact the Head of Regulatory Compliance at:

Research Integrity and Governance Team
Imperial College London and Imperial College Healthcare NHS Trust
Room 215, Level 2, Medical School Building
Norfolk Place
London, W2 1PG
Tel: **0207 594 9459/ 0207 594 1862**
http://www3.imperial.ac.uk/clinicalresearchgovernanceoffice

This protocol describes the PREGRRS study and provides information about procedures for entering participants. Every care was taken in its drafting, but corrections or amendments may be necessary. These will be circulated to investigators in the study. Problems relating to this study should be referred, in the first instance, to the Chief Investigator.

This study will adhere to the principles outlined in the UK Policy Frame Work for Health and Social Care Research. It will be conducted in compliance with the protocol, the Data Protection Act and other regulatory requirements as appropriate.

GLOSSARY OF ABBREVIATIONS

TSH	Thyroid Stimulating Hormone
fT4	Free thyroxine
fT3	Free triiodothyronine

KEYWORDS

Thyroid, Pregnancy, Women, Reference Range, Laboratory, Blood Test

STUDY SUMMARY

TITLE	Pregnancy Reference Range
DESIGN	Interventional, cross-sectional study
AIMS	To produce accurate, trimester-specific reference ranges for plasma and urine biochemistry in pregnancy
OUTCOME MEASURES	Laboratory blood test results
POPULATION	n=1200
ELIGIBILITY	Pregnant women receiving antenatal care at Queen Charlottes' Hospital
DURATION	1 hour per participant, 6 months total

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1. INTRODUCTION

1.1 Background

A woman's physiology changes dramatically once she becomes pregnant. Included in this are changes in many biochemical and haematological measures. These include increased production of clotting factors and elevated levels of D-dimers; a fall in plasma potassium and sodium levels, and a mild increase in white cells (Abbassi-Ghanavati, Greer, & Cunningham, 2009).

Physicians and obstetricians need pregnancy-specific reference ranges for blood markers to offer optimal care for pregnant women and the developing foetus. For example, maternal thyroid hormone crosses the placenta and affects fetal neuro-development. However, beta-hCG and oestrogen produced by the placenta have dramatic effects on the maternal thyroid and associated hormones throughout pregnancy (see figure 1 below). Therefore trimester-specific reference ranges are needed to ensure that women can be diagnosed in pregnancy with an over-or under-active thyroid, and treated appropriately whether they have a new diagnosis or known pre-existing thyroid dysfunction. However, widely available reference ranges for many blood markers, including thyroid function, does not exist or have not been updated as we have adopted new methods of measuring the analytes.

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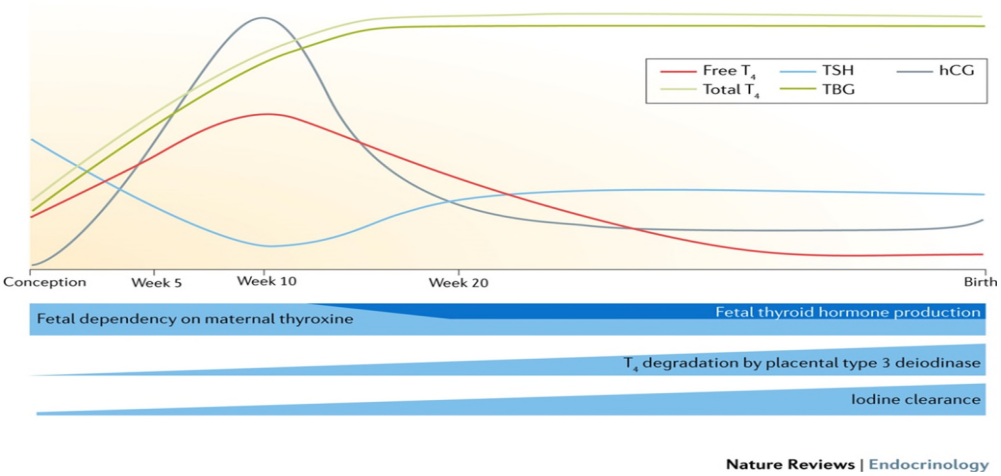


Figure 1: from Korevaar, T. I. M. *et al.* (2017) Thyroid disease in pregnancy: new insights in diagnosis and clinical management *Nat. Rev. Endocrinol.* doi:10.1038/nrendo.2017.93

1.2 Rationale for current study

The primary aim of this study, in line with the recommendations from the American Thyroid Association 2017 guidelines for the management of thyroid disease in pregnancy, is to produce trimester-specific thyroid reference ranges to be used within the Imperial College Healthcare NHS Trust. ICHNT includes two maternity units which cover approximately 9000 childbirths per year. Local audit has shown that thyroid function tests are checked in approximately 15% of these women, despite no accurate trimester-specific reference ranges being available. Recently, new biochemical analysers have been installed at ICHNT (Abbot Alinity). By producing accurate reference ranges we will be able to offer better care for all these women. Furthermore, as the population served by ICHNT is ethnically and culturally diverse, these reference ranges will be transferrable to other hospitals using the Alinity platform. If there is sufficient sample volume, we will also run these samples on other biochemical assay platforms, such that these reference ranges can be applicable to a wider range of settings. In addition, we will measure urinary iodine levels as iodine levels affect thyroid function and recent studies have suggested that women in the UK are iodine deficient (Vanderpump et al,2011).

In addition to analysing thyroid hormones, we will store samples to allow subsequent analysis and production of trimester-specific reference ranges for other biochemical markers. Pregnant women are rarely included in clinical studies, and there is a significant gap in reference ranges for pregnant women which lead to suboptimal clinical care. Therefore we would like to optimise the use of these precious samples by studying other analytes. These will include troponin, a marker of heart disease, and the blood pressure hormones renin and aldosterone. ICHNT is the supra-regional assay service for the measurement of aldosterone and renin. Samples are sent to ICHNT from throughout the UK when there are queries about pregnant women with rare but serious medical conditions such as Conn's disease (which increases aldosterone levels). By producing contemporary reference ranges for these analytes, more accurate care can be offered to women across the country with rare but serious conditions in pregnancy. Troponin is a marker of cardiac muscle disease. As cardiac disease is a leading cause of maternal death (MBBRACE report 2019, https://www.npeu.ox.ac.uk/assets/downloads/mbrrace-uk/reports/MBRRACE-UK%20Maternal%20Report%202019%20-%20Infographic%20v1.0.pdf) being able to accurately and promptly diagnose maternal heart disease throughout pregnancy will save maternal lives.

2. STUDY OBJECTIVES

Primary Objective

- To establish trimester-specific reference ranges for thyroid axis hormones in pregnancy

Secondary Objective

- To establish trimester-specific reference ranges for other plasma and urine analytes in pregnancy such as renin, aldosterone and troponin.
- To determine urinary iodine concentrations in the pregnant population in London.

3. STUDY DESIGN

Interventional cross-sectional study in pregnant women

1 hour per participant, up to 2 years to complete.

n= 400 women in each trimester: to achieve 400 blood samples in each trimester and 100 urine samples in each trimester.

3.1 Study Outcome Measures

Trimester-specific ranges for plasma and urine biochemistry

4. PARTICIPANT ENTRY

4.1 Pre-registration evaluations

Potential participants will be identified by the antenatal teams from all clinic, phlebotomy and ultrasound lists of women attending Queen Charlotte and Chelsea Hospital (QCCH) for antenatal care.

4.2 Inclusion Criteria

- Women >18 years of age who are receiving antenatal care at QCCH
- Singleton pregnant

4.2 Exclusion Criteria

- Multiple pregnancy
- Current or previous history of thyroid disorder
- Previous or current thyroid medication use
- Inability to understand and write in the English language
- Unable to participate due to other factors, as assessed by the Chief Investigators

4.3 Withdrawal criteria

- Loss of capacity to give informed consent
- Investigator initiated discontinuation of study due to participation concerns

Withdrawal will be immediate, and participants will be followed up with appropriate antenatal care.

If a participant, who has given informed consent, loses capacity to consent during the study the participant will be withdrawn from the study. Identifiable data already collected with consent will be

retained and used in the study. No further data would be collected or any other research procedures carried out on or in relation to the participant.

5. ADVERSE EVENTS

5.1 Definitions

Adverse Event (AE): any untoward medical occurrence in a patient or clinical study subject.

Serious Adverse Event (SAE): any untoward and unexpected medical occurrence or effect that:

- **Results in death**
- **Is life-threatening** – *refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe*

- **Requires hospitalisation, or prolongation of existing inpatients' hospitalisation**
- **Results in persistent or significant disability or incapacity**
- **Is a congenital anomaly or birth defect**

Medical judgement 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 also be considered serious.

5.2 Reporting Procedures

All adverse events should be reported. Depending on the nature of the event the reporting procedures below should be followed. Any questions concerning adverse event reporting should be directed to the Chief Investigator in the first instance.

5.2.1 Non serious AEs

All such events, whether expected or not, should be recorded.

5.2.2 Serious AEs

An SAE form should be completed and faxed to the Chief Investigator within 24 hours. However, hospitalisations for elective treatment of a pre-existing condition do not need reporting as SAEs.

All SAEs should be reported to the Cambridge South Research Ethics Committee where in the opinion of the Chief Investigator, the event was:

- 'related', i.e. resulted from the administration of any of the research procedures; and
- 'unexpected', i.e. an event that is not listed in the protocol as an expected occurrence

Reports of related and unexpected SAEs should be submitted within 15 days of the Chief Investigator becoming aware of the event, using the NRES SAE form for non-IMP studies. The Chief Investigator must also notify the Sponsor of all SAEs.

Local investigators should report any SAEs as required by their Local Research Ethics Committee, Sponsor and/or Research & Development Office.

Contact details for reporting SAEs
jrco@imperial.ac.uk
CI email (and contact details below)
Fax: 0202 312 1563 for attention Dr Rebecca Scott
Please send SAE forms to: Department of Diabetes and Endocrinology, Hammersmith Hospital, Du Cane Road, London, W12 0NN
Tel: 0203 312 1253 (Mon to Fri 09.00 – 17.00)

6. ASSESSMENT AND FOLLOW-UP

This study will consist of a questionnaire, and taking no more than 20ml blood samples and a 100ml urine sample from women attending for routine antenatal care.

All eligible participants will be identified by the clinical care team. Potential participants will be identified from women attending for routine antenatal clinics, antenatal scans and antenatal phlebotomy. Women will be approached by the clinical team about participating in the study. The contact details of women who are interested will be passed to the research team. The research team will then contact the woman by phone or email, and send the woman the patient information sheet, either by email or by post. Women will then have up to the end of their pregnancy to read the PIS and agree to participate in the study.

Women who then agree to participate will be asked to fill in an electronic consent form. To do this they will be sent a link to Qualtrics (an online data collection platform with is supported by Imperial College). They will also be asked to complete a short questionnaire also via a link to Qualtrics. Both links will be individualised, and the questionnaire data will be pseudonymised and linked to their unique study number. The study number will enable the research team to link the questionnaire data and the samples taken

The questionnaire will contain basic details about the woman and her pregnancy including

- Date of birth
- Weight pre- and post-pregnancy
- Height
- Ethnicity
- Estimated date of delivery and Last menstrual period
- Medical history including thyroid disease and medication history
- Use of a pregnancy multivitamin

After they have received the consent form and questionnaire, participants will have up to 72 hours prior to any face-to-face antenatal visit to decide to participate and complete the consent form.

The blood and urine samples will be taken by either the research team, or by the routine phlebotomy service at Queen Charlotte & Chelsea Hospital. These will be taken at any point that the woman attends the antenatal service. The samples labelled with the unique study number, but no

other data related to the individual will be found on the samples.

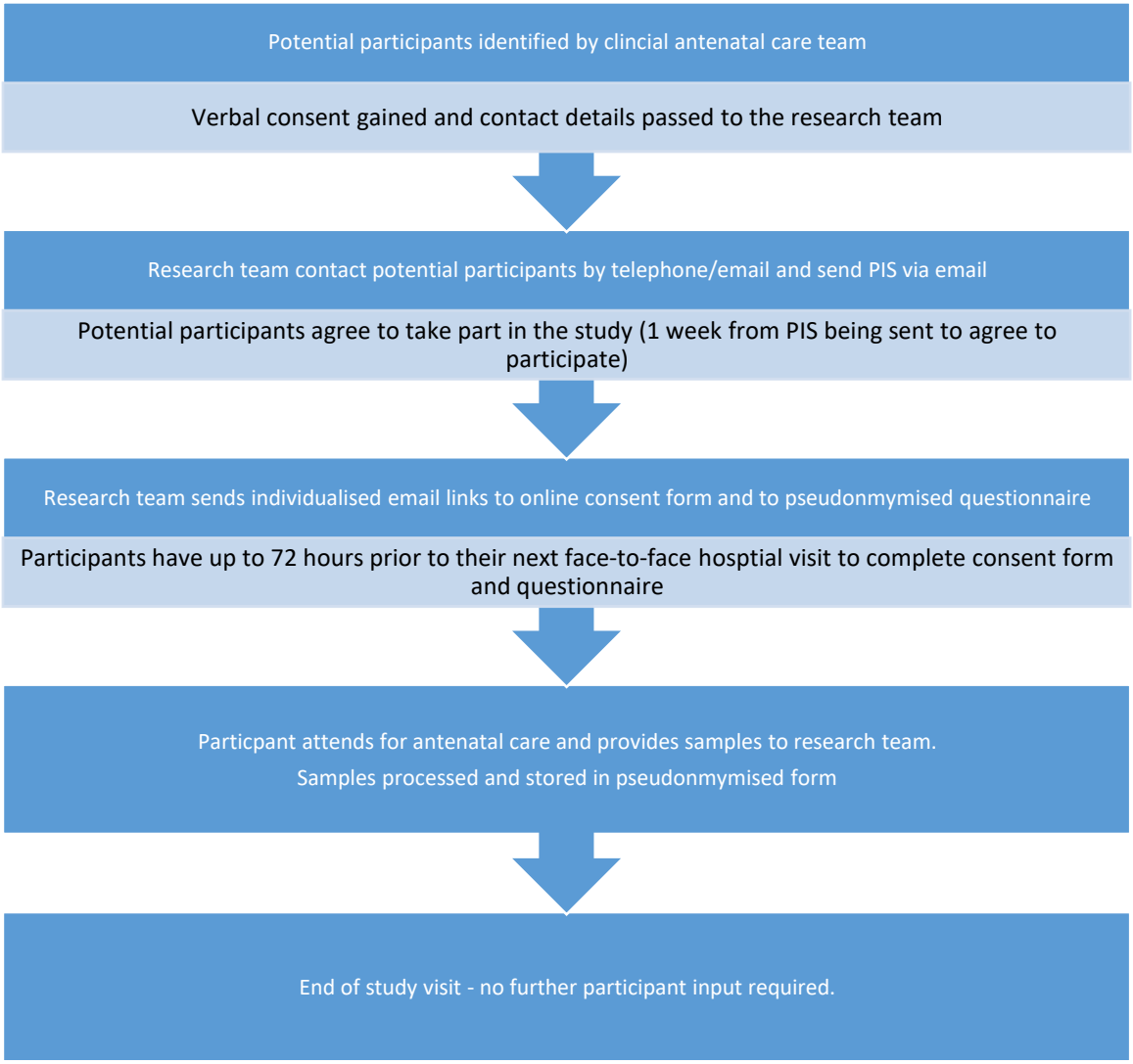
The samples will then be delivered to the Imperial College Healthcare NHS Trust clinical laboratories. The samples taken will be aliquoted. Two aliquots, pseudo-anonymised with the unique study number, will be processed within the laboratory. The results will be recorded in the pseudonymised form in the results database. The other aliquots will be securely stored, in the pseudonymised form, within specific study freezers at ICHNT.

Initially blood samples will be processed to determine TSH, fT4 and fT3 levels, plus TPO antibody levels, while urine samples will be tested for iodine concentration. Stored samples will be used for other biochemical markers such as aldosterone, renin and troponin.

The study database will be securely held on ICHNT computers.

If abnormal results are found (TSH >10 mU/l in any trimester, or TSH <0.03 in the 2nd and 3rd trimesters) the woman will be contacted and the results discussed. Otherwise no follow up outside of usual care will be provided.

End of study will be defined as last subject last visit (LSLV).


Potential participants identified by clincial antenatal care team
Verbal consent gained and contact details passed to the research team
Research team contact potential participants by telephone/email and send PIS via email
Potential participants agree to take part in the study (1 week from PIS being sent to agree to participate)
Research team sends individualised email links to online consent form and to pseudonmymised questionnaire
Participants have up to 72 hours prior to their next face-to-face hosptial visit to complete consent form and questionnaire
Particpant attends for antenatal care and provides samples to research team.
Samples processed and stored in pseudonmymised form
End of study visit - no further participant input required.

7. STATISTICS AND DATA ANALYSIS

As per the Association of Clinical Biochemists, as TSH is skewed in the normal population, TSH reference intervals will be established from the 95 % confidence limits of the log-transformed values of at least 120 rigorously screened normal euthyroid volunteers who are TPO antibody negative. From local preliminary data, 4% of women had TPO antibodies; national data suggests approximately 15% of women of childbearing age have TPO antibodies (Dhillon-Smith et al., 2019). Therefore 150 women in each trimester should provide rigorous reference ranges. However as thyroid function is also influenced by ethnicity, we will aim to recruit up to 400 per trimester to ensure representation of minority ethnic groups.

All other analytes (e.g. fT4 andfT3) are normally distributed, reference ranges will be established from 95% confidence limits of the samples taken. They will be reported as per national or international society guidelines.

Data and all appropriate documentation will be pseudonymised and stored for a minimum of 10 years after the completion of the study.

8. REGULATORY ISSUES

8.1 Ethics approval

The Study Coordination Centre has obtained approval from the Cambridge South Research Ethics Committee (REC) and Health Regulator Authority (HRA). The study must also receive confirmation of capacity and capability from each participating NHS Trust before accepting participants into the study or any research activity is carried out. The study will be conducted in accordance with the recommendations for physicians involved in research on human subjects adopted by the 18th World Medical Assembly, Helsinki 1964 and later revisions.

8.2 Consent

Consent to enter the study must be sought from each participant only after a full explanation has been given, an information leaflet offered and time allowed for consideration. Signed participant consent should be obtained. Consent may be gained electronically via a link to Qualtrics, a secure online data collection tool supported by Imperial College. The right of the participant to refuse to participate without giving reasons must be respected. After the participant has entered the study the clinician remains free to give alternative treatment to that specified in the protocol at any stage if he/she feels it is in the participant's best interest, but the reasons for doing so should be recorded. In these cases the participants remain within the study for the purposes of follow-up and data analysis. All

participants are free to withdraw at any time from the protocol treatment without giving reasons and without prejudicing further treatment.

8.3 Confidentiality

The Chief Investigator will preserve the confidentiality of participants taking part in the study and is registered under the Data Protection Act.

8.4 Indemnity

Imperial College London holds negligent harm and non-negligent harm insurance policies which apply to this study.

8.5 Sponsor

Imperial College London will act as the main Sponsor for this study. Delegated responsibilities will be assigned to the NHS trusts taking part in this study.

8.6 Funding

The study is funded by a pump-priming grant from North West London Pathology, and by contract with Abbott Diagnostics. No payments will be made to participants or investigators in this study.

8.7 Audits

The study may be subject to inspection and audit by Imperial College London under their remit as sponsor and other regulatory bodies to ensure adherence to GCP and the UK Policy Frame Work for Health and Social Care Research.

8.8 International Transfer

There may be a requirement to transfer information to countries outside the European Economic Area (for example, to a research partner). Where this information contains your personal data, Imperial College London will ensure that it is transferred in accordance with data protection legislation. If the data is transferred to a country which is not subject to a European Commission (**EC**) adequacy decision in respect of its data protection standards, Imperial College London will enter into a data sharing agreement with the recipient organisation that incorporates EC approved standard contractual clauses that safeguard how your personal data is processed. In this study, the study protocol and results will be shared with the study funder Abbott Diagnostics; however all shared data will be anonymised.

9. STUDY MANAGEMENT

The day-to-day management of the study will be co-ordinated through Dr Rebecca Scott.

10. PUBLICATION POLICY

The study will be registered on the ISRCTN registry system and results will be disseminated by peer reviewed scientific journals, internal report, conference presentation and publication on websites. No identifiable personal data will be published. All anthropometry and personal clinical data will be expressed as mean/ median and spread of the population in the study.