Evaluation of Digital Microfluidic Molecular Point of Care Testing for Diagnosis of Respiratory Pathogens

Short Title: **S**tudy for **M**ultiplex **A**ssessment and **R**espiratory **T**est Evaluation; **SMART**

Version 1.0

Dated 09/OCT/2025

IRAS Number: 363005

Sponsor Protocol Reference Number: SMARTUK01

Sponsor:

Logilet (UK) Limited

Flat 107

25 Indescon Square

London

E14 9DG

United Kingdom

1.0 SIGNATURE PAGE

The undersigned confirm that the following protocol has been agreed and accepted and that the Chief Investigator agrees to conduct the study in compliance with the approved protocol and will adhere to the principles outlined in the Declaration of Helsinki, the Sponsor's SOPs, and other regulatory requirements.

I agree to ensure that the confidential information contained in this document will not be used for any other purpose other than the evaluation or conduct of the investigation without the prior written consent of the Sponsor.

I also confirm that I will make the findings of the study publicly available through publication or other dissemination tools without any unnecessary delay and that an honest, accurate and transparent account of the study will be given; and that any discrepancies from the study as planned in this protocol will be explained.

Sponsor Representative Signature

Signature:	Date:
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Name (please print):	
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Position:	
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Chief Investigator Signature

Signature:	Date:
....	....../....../......
Name (please print):	
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Position:	
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2.0 CONTENTS LIST

3.0 STUDY CONTACT INFORMATION

Sponsor Representative	Name: Dr Ruichao Bai (Barry) Address: Flat 107, 25 Indescon Square, London, E14 9DG Email: ruichao.bai@logilet.com Phone: +86 18600138212
Chief Investigator	Name: Dr Adrian Boyle Address: Emergency Department, Box 87, Addenbrooke's Hospital, Cambridge Biomedical Campus, Hills Road, Cambridge, CB2 0QQ Email: adrianboyle@nhs.net Phone: 01223 596145
Research Team	Address: Emergency Department, Box 87 Addenbrooke's Hospital, Cambridge Biomedical Campus, Hills Road, Cambridge, CB2 0QQ Email: cuh.edresearchteam@nhs.net Phone: 01223 217907

4.0 SUMMARY OF PROTOCOL AMENDMENTS

Summary of Protocol Amendments (Substantial and Non-Substantial)			
Amendment Number	New Protocol Version and Date	Description of Change	Reason for Change
NA	NA	NA	NA

5.0 PROTOCOL SYNOPSIS

Study Summary	
Protocol Title	Evaluation of Digital Microfluidic Molecular Point Of Care Testing for Respiratory Pathogens Diagnosis
Short Title	**S**tudy for **M**ultiplex **A**ssessment and **R**espiratory **T**est evaluation; **SMART**
Study Objectives	To evaluate the analytical performance, turnaround time, operability and acceptability of a novel multiplex respiratory pathogens point of care test device compared to currently available point of care testing.
Study Procedures	Participants will be swabbed to provide samples which will be analysed by standard testing and by the novel multiplex device. The procedures will take place in a single visit in the Emergency Department at Addenbrooke's Hospital in Cambridge. Additional relevant data will be collected from the participants' hospital medical records. No further participant involvement is required.
Study Sponsor	Logilet (UK) Ltd
Sponsor Representative	Mr Ruichao Bai (Barry) Product Manager
Study Design	This is a prospective, single centre, single visit, diagnostic performance study. There is no therapeutic agent involved and therefore no randomisation into treatment arms. Two nasopharyngeal swabs will be taken simultaneously from each participant (one from either nostril) and used for Point Of Care Testing using the current gold standard and novel *in vitro* diagnostic devices. In this way, participants will act as their own controls for comparison of results. If the initial standard test is negative but the participant continues to exhibit clinical symptoms of respiratory infection, a further swab may be taken and tested as per Cambridge University Hospitals NHS Foundation Trust (CUH) policy with another standard device.
Investigational Medical Device	The Logicore System *in vitro* diagnostic device utilises digital microfluidics to rapidly test for the presence of 6 respiratory pathogens, namely SARS-CoV-2, Influenza A (FluA), Influenza

	B (FluB), Respiratory Syncytial Virus (RSV), *Mycoplasma pneumoniae* (MP) and human adenovirus (HAdV). The Logicore System produces results in approximately 63 minutes and is expected to feature improved sensitivity when compared to the current gold standard testing device.
Study Location	Cambridge University Hospitals NHS Foundation Trust (CUH) Emergency Department Addenbrooke's Hospital Cambridge Biomedical Campus Hills Road Cambridge CB2 0QQ
Study Duration	When the sample size has been reached, or a maximum of four months from the time of first participant enrolled to the last subject recruited into the study.
Subject Population	Male and female participants, of any age, meeting clinically suspected respiratory tract infections case criteria, with a signed informed consent form, and who require standard of care diagnostic testing as per CUH Trust Infection Prevention and Control policy.
Sample Size	400 participants
Primary Endpoint	Demonstration of the sensitivity and accuracy of the new device compared to current gold standard testing to include Positive Predictive Value (PPV), Negative Predictive Value (NPV) and sensitivity.
Secondary Endpoints	1) Comparison of the turnaround time for the new device compared to gold standard point of care testing. 2) Evaluation of acceptability of the new device by study participants and staff. 3) Targeted Cost Benefit analysis of the new device.
Reference Standards	● UK Medical Devices Regulation 2002 (SI 2002 No 618, as amended) ● In Vitro Diagnostic Device Regulation (EU) 2017/746

	• ISO 20916:2019 In vitro diagnostic devices - Clinical performance studies using specimens from human subjects – good study practice • Declaration of Helsinki 2024 – Medical Research Involving Human Participants • Declaration of Helsinki 2008 – Ethical Principles for Medical Research • ICH Guideline E6(R2) 2016: Good Clinical Practice • ICH Guideline E8: General Considerations for Clinical Trials • ICH Guideline E9: Statistical Principles for Clinical Trials
Health Economics	The time taken to diagnose a patient presenting at the Emergency Department (ED) and initiate appropriate treatment can influence the patient's outcome and disposition. The quicker the therapeutic regime is started, the better outcome for the patient. Furthermore, the sooner the presence of a pathogen is confirmed, the quicker the patient can be appropriately isolated to stem the spread of infection. Current diagnostic testing via the local hospital laboratory (remote to the ED) can take up to an hour and a half from time of swabbing the patient, to receipt of test results. The novel Logicore System device can be used in the ED at the Point of Care (a formal laboratory setting is not necessary) and provides a result in approximately one hour. This device could cut diagnosis time and therefore time to initiation of appropriate treatment, provide better infection prevention control (by identification of those patients that need and do not need isolation measures), streamline the patient treatment pathway and potentially improve outcomes for patients with respiratory infections whilst reducing spread of infection within the hospital. Adopting the Logicore System testing device could benefit patients whilst also benefitting the Trust in terms of reduced economic burden especially in times of winter pressures.
Randomisation	There is no treatment or intervention in this study and therefore no randomisation. Specimens from each participant will be tested using gold standard means and the novel device.

Eligibility Criteria	**Inclusion Criteria** 1) Male or female 2) Any age 3) Presenting to CUH Emergency Department 4) Symptomatic of respiratory tract infection by clinical evidence of any of the following: • Acute respiratory distress syndrome • Influenza like illness • Fever ≥ 37.8°C • Acute onset persistent cough (with or without sputum), hoarseness, nasal discharge or congestion, shortness of breath, sore throat, wheezing or sneezing • Any other symptom known to be indicative of acute respiratory episode 5) Signed consent form for participation 6) Requiring standard of care diagnostic testing as per CUH Trust Infection Prevention and Control policy 7) Able to read and/or understand the age-appropriate participant information sheet in English. **Exclusion Criteria** 1) Unwilling or unable to comply with study nasopharyngeal swabbing procedures 2) Those who are incapacitated or deemed to be lacking capacity to provide informed consent to participate 3) Prisoners or young offenders.

6.0 ACRONYMS AND DEFINITIONS

Acronym	Definition
ADE	Adverse Device Effect
AE	Adverse Event
APR	Annual Progress Report
CAP	College of American Pathologists
CDC	United States Centers for Disease Control and Prevention

CE	Conformitié Européenne
CEA	Cost Effectiveness Analysis
CI	Confidence Interval
COVID-19	Coronavirus Disease of 2019
CTCAE	Common Terminology Criteria for Adverse Events v5.0
CUH	Cambridge University Hospitals NHS Foundation Trust
DD	Device Deficiency
DPO	Data Protection Officer
ECDS	Emergency Care Data Set
eCRF	Electronic Case Report Form
ED	Emergency Department
ERS	European Respiratory Society
EU	European Union
FluA	Influenza A
FluB	Influenza B
GCP	Good Clinical Practice
GDPR	General Data Protection Regulation
HAdV	Human adenovirus
HRA	Health Research Authority
HSA	Health Security Agency
ICER	Incremental Cost Effectiveness Ratio
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
IFU	Instructions For Use
IPC	Infection Prevention and Control
IRAS	Integrated Research Application System
ISF	Investigator Site File
ISO	International Organization for Standardization
ITT	Intention To Treat
IVDD	In Vitro Diagnostic Device
IVDR	In Vitro Device Regulation (EU) 2017/746
LOS	Length of Stay
mCTA	model Clinical Trial Agreement
MDR 2002	Medical Devices Regulations 2002 (SI 2002 No 619, as amended)
MHRA	Medicines and Healthcare products Regulatory Agency
MP	*Mycoplasma pneumoniae*
NA	Not Applicable
NCVR	National Contract Value Review
NEWS-2	National Early Warning Score 2
NHS	National Health Service
NIHR	National Institute for Health and Care Research
NPV	Negative Predictive Value
PCR	Polymerase Chain Reaction
PEWS	Paediatric Early Warning Score
PID	Participant Identifiable Data

PIS	Participant Information Sheet
POC	Point Of Care
POCT	Point Of Care Testing
PPE	Personal Protective Equipment
PPV	Positive Predictive Value
PRC	People's Republic of China
QALY	Quality-Adjusted Life Year
qPCR	Quantitative Polymerase Chain Reaction
REC	Research Ethics Committee
RN	Research Nurse
RSV	Respiratory Syncytial Virus
RT-PCR	Reverse Transcription Polymerase Chain Reaction
SADE	Serious Adverse Device Effect
SAE	Serious Adverse Event
SARS	Severe Acute Respiratory Syndrome
SD	Standard Deviation
SDV	Source Data Verification
SIV	Site Initiation Visit
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reaction
TMF	Trial Master File
UK	United Kingdom
USADE	Unanticipated Serious Adverse Device Effect
WHO	World Health Organisation

7.0 INTRODUCTION

7.1 Background

Respiratory tract infections are the commonest cause of morbidity and mortality amongst acute infectious diseases[1]. The spread of respiratory diseases is quick and poses challenges especially in hospital settings with potentially vulnerable patients. It is, therefore, imperative to expedite diagnosis to facilitate correct and appropriate triage and infection control measures. Rapid turnaround of diagnosis of multiple, frequently occurring respiratory infections also ensures the appropriate treatment and appropriate Infection Prevention and Control (IPC) measures are instituted early upon presentation and detection.

It has been of paramount importance to develop and evaluate diagnostic tests during the COVID-19 pandemic for many reasons; firstly, to diagnose infected cases, so they may be treated appropriately and secondly, to identify cases to quarantine and stop nosocomial transmission. The point-of-care (POC) molecular diagnostic tests have radically improved and changed the way we address the management of these cases.

The standard diagnostic Point Of Care Testing (POCT) by polymerase chain reaction (PCR) usually detects at most 4 respiratory pathogens. Additional PCR testing and United Kingdom Health Security Agency (UK HSA) laboratory analysis may be required, causing obvious bottlenecks, and extended turnaround time. A rapid multiplex POC test is very much needed, that will detect a broader range of respiratory pathogens whilst maintaining a quick turnaround of results, without trading off sensitivity and specificity.

The CUH IPC Guideline[2] states all symptomatic patients who are to be admitted from ED must have Cepheid monoplex POCT PCR assay performed to test for SARS-CoV-2. During the winter months when there is a high prevalence of seasonal respiratory viruses, a Cepheid multiplex POCT PCR assay is utilised instead which tests for influenza A, influenza B, Respiratory Syncytial Virus (RSV) and SARS-CoV-2. Hospital inpatients who did not exhibit respiratory symptoms at admission but go on to develop them are tested using a SARS-CoV-2 POCT PCR (SAMBA) test and also UK HSA laboratory respiratory PCR swab Luminex analysis which tests for human adenovirus, seasonal coronaviruses, human bocavirus, human metapneumovirus, influenza A (H1, H3, 2009 H1N1), influenza B, parainfluenza (1-4), picornaviruses (rhino/ enterovirus), RSV A and B, *Chlamydia pneumoniae*, *Legionella pneumophila* and *Mycoplasma pneumoniae*. Patients who are admitted to hospital after testing negative on the Cepheid POCT in the ED but with ongoing clinical concern for respiratory viral infection, may also undergo additional UK HSA laboratory respiratory virus PCR testing.

This study aims to recruit participants during the winter months, when the gold standard POCT device used at CUH is the Cepheid multiplex PCR assay (known as GeneXpert). This test can produce results in around 40 minutes to an hour. The advantage of utilising the Logicore System is that it can test for 2 additional pathogens in roughly the same amount of time as Cepheid (it takes 63 minutes to produce the results) but is expected to be more sensitive to the pathogens (for example, the minimum limit of detection is 131 copies/mL for SARS-CoV-2 using Cepheid[3] but is expected to be only 100 copies/mL for the same pathogen using the Logicore System). This could alleviate the need for additional UK HSA laboratory testing if a patient was admitted following a negative Cepheid test but continued to exhibit respiratory symptoms, as it is more likely the pathogen would be identified in the initial Logicore System testing process. The CUH IPC Guideline instructs that patients with suspected respiratory infection should be isolated while further UK HSA lab PCR testing is performed. Therefore, the improved sensitivity in diagnostic capability could reduce the need for unnecessary isolation of patients as there would be reduced requirement for the secondary HSA testing. Conversely, the improved turnaround time would ensure rapid instigation of appropriate isolation procedures when a patient has positive initial results to reduce the spread of infection in the ED, in conjunction with quicker treatment of patients with targeted therapeutics medicinal products.

This study aims to produce evidence to support the Logicore System claims of increased sensitivity and specificity, whilst maintaining accuracy, and reduced turnaround times compared to current practice, and to provide the Trust with a Cost Benefit analysis from assessment with the Logicore System.

7.2 The Investigational Medical Device

The Investigational Medical Device being assessed in this clinical study is an *in vitro* diagnostic device (IVDD) that has been developed and manufactured by Nanjing Vazyme Biotechnology Company Limited (Vazyme) in the People's Republic of China (PRC). It has already achieved CE marking status in the EU. The CE mark denotes that the device has been produced in accordance with the health, safety and environmental requirements to be sold in the European Economic Area. Logilet (UK) Ltd is the UK subsidiary of Vazyme.

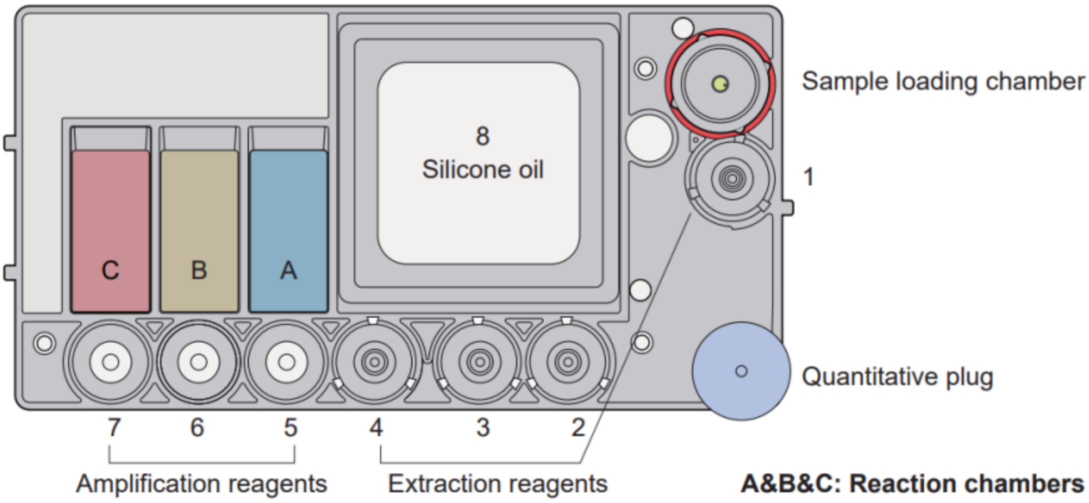
The Logicore System has been certified by the College of American Pathologists (CAP) with 100% detection accuracy for the 6 respiratory pathogens SARS-CoV-2, Influenza A (FluA), Influenza B (FluB), Respiratory Syncytial Virus (RSV), *Mycoplasma pneumoniae* (MP) and human adenovirus (HAdV)].

The Logicore System comprises a portable Operation Module with display screen, an Analytical Module and a Respiratory Pathogen Panel cartridge as shown overleaf:

The Respiratory Pathogen Panel containing digital microfluidics is a Class D *in vitro* diagnostic device as defined in the EU In Vitro Device Regulation (IVDR) 2017/746[4].

The Respiratory Pathogen Panel is inserted into the Analytical Module, and the results are displayed on the portable Operation Module following analysis.

The Respiratory Pathogen Panel cartridge is shown below:



The swab to be tested for presence of respiratory pathogens is placed in a Preservation Buffer (provided to the site by Logilet) and a droplet is then placed in the sample loading chamber of the Respiratory Pathogen Panel cartridge. The Preservation Buffer is stable for 12 months at ambient storage. One Operation Module can be connected to up to 8 Analytical Modules and one cartridge is used per sample. Thus, in this scenario 8 samples can be analysed in approximately one hour. For this clinical study, the site will initially be provided with 2 Operation Modules, to enable 2 Respiratory Pathogen Panel cartridges to be analysed in a one-hour period. If there is sufficient space in the ED, and once the staff are familiar with using the Logicore System, a further 2 Operation Modules (maximum of 4 in total) may be provided to allow 4 cartridges to be analysed at once (i.e. samples from 4 participants).

The Operation Modules are designed to be safely stacked as shown overleaf:

8.0 RISK/BENEFIT ANALYSIS

8.1 Anticipated Risks

During the development and CE marking process, no new unexpected risks were identified for staff using, or participants being tested with, the Logicore System. Whenever a nasopharyngeal swab is used, there is a potential risk of discomfort and/or a small possibility of nosebleed. This will be explained to the participants during the informed consent process and staff are trained in the swabbing procedure to minimise the likelihood of discomfort and/or nosebleeds. The ED research nurse (or designated member of the research team) will wear standard Personal Protective Equipment (PPE) and follow standard measures for cleaning and disinfection to reduce the potential risk of spreading infection between staff and patients, in line with the CUH Infection Control and Prevention Guideline. Furthermore, the Logicore System testing cartridge is fully sealed and the PCR reaction is performed in a silicone oil medium. This double feature minimises risk of contamination and prevents PCR aerosol pollution and bio-hazard risks.

The research staff will be trained to use the Logicore System during the Site Initiation process and will be provided with Instructions For Use to minimise the risk of the device being used incorrectly. Staff will be prompted to check expiry dates to ensure test components are in date and have been stored according to the Sponsor requirements.

8.2 Potential Benefits

The main potential benefits from using the Logicore System device compared to standard testing means are as follows:

1) The Logicore System can test for 6 different respiratory pathogens all at once, compared to 4 for the Cepheid GeneXpert device.

2) The Logicore System is expected to be more sensitive than the standard device. The minimum limit of pathogen detection is 100 copies/mL whereas the Cepheid GeneXpert device has a minimum limit of detection of 131 copies/mL for SARS-CoV-2[3].

3) The Logicore System produces rapid results in around one hour, compared to the hour and a half using the UK HSA laboratory second-line test if the initial Cepheid test is negative but clinical symptoms persist.

There is no particular benefit to individual participants from taking part in the study. There are no payments to participants for taking part and the study has no bearing on the clinical care each participant receives. Once the swabs have been taken from participants, they will immediately return to standard clinical care without delay. However, if the Logicore System testing device detects a pathogen that is not identified by the Cepheid test, it may result in faster commencement of treatment with an appropriate therapeutic drug regime. Instigation of treatment will take place outwith the study and will be undertaken by the participant's ED care team. There is no Investigational Medicinal Product in the study itself.

Additionally, participants may experience a sense of altruism from taking part in a study aiming to reduce diagnostic time and spread of infection for future ED patients.

Any risks have been mitigated such that it is considered the benefits to those taking part in the study outweigh any potential risks.

9.0 STUDY OBJECTIVES

9.1 Primary Objective

The primary objective of the study is to demonstrate that the Logicore System multiplex 6 panel respiratory pathogens point of care testing device has increased diagnostic sensitivity when compared to the current gold standard POC multiplex panel at CUH Trust whilst maintaining accuracy, including Positive Predictive Value (PPV), Negative Predictive Value (NPV) and specificity.

9.2 Secondary Objectives

1) To compare the time from sample acquisition to receipt of result (turnaround time) for Logicore System point of care testing and current POC testing (Cepheid multiplex GeneXpert)

2) To evaluate the acceptability of the Logicore System device by study participants and staff.

3) To produce targeted Cost Benefit analysis for the Logicore System device.

10.0 STUDY ENDPOINTS

10.1 Primary Endpoint / Outcome Measure

The primary endpoint of the study is to measure the sensitivity and accuracy of Logicore System POC diagnostic test compared to gold standard POC multiplex test (Cepheid GeneXpert), including PPV, NPV and specificity.

10.2 Secondary Endpoint / Outcome Measures

1) Determination of turnaround time for Logicore System and Cepheid tests (time taken from nasopharyngeal swabbing procedure to receipt of positive result).

2) Evaluation of acceptability of Logicore System POCT to study participants and staff.

3) Targeted Cost Benefit analysis of Logicore System POCT.

11.0 STUDY DESIGN

This is a prospective, single-centre, diagnostic accuracy study, being conducted at Addenbrooke's Hospital in Cambridge. Participants can be any age but must exhibit acute respiratory symptoms on presentation at the Emergency Department and require standard of care diagnostic testing as per CUH Trust IPC policy.

The study will be performed in accordance with the Declaration of Helsinki[5], principles of Good Clinical Practice[6] and local and regulatory requirements.

The study is aiming to recruit 400 participants. There is no Investigational Medicinal Product involved and therefore no treatment arms and no randomisation. Swab samples taken from each participant will be tested by standard POCT (control data) and by Logicore System POCT device.

Members of the research ream will utilise Personal Protective Equipment (PPE) in line with the CUH Infection Prevention and Control Guideline.

There is only one visit required for each participant, during which the following data will be collected:

- Study identifying number
- Date of Visit and Study Assessments (presentation at ED)
- Sex
- Age in years
- Emergency Care Data Set (ECDS) presentation code
- Clinical signs and symptoms of respiratory tract infection
- Date of onset of clinical symptoms of respiratory tract infection
- Time nasopharyngeal swabs taken
- Time results available from Cepheid and Logicore System devices (to establish turnaround time in minutes)
- Hospital admission required? Y/N
- Intensive care admission required? Y/N
- NEWS-2 Score for participants 15 years of age and above
- PEWS (Paediatric Early Warning Score) for participants less than 15 years of age
- Arrival by ambulance? Y/N
- Respiratory rate (breaths per minute)
- Temperature (in °C)
- Heart rate (beats per minute)
- Blood pressure (systolic and diastolic)
- Oxygen saturation (%)
- Confirmation that Logicore System Instructions For Use (IFU) were followed to test the sample using the new device
- Participant and staff assessment of acceptability
- Adverse event details.

Following discharge from the ED (to home, nursing home or a hospital ward etc), the following additional information will be collected:

- 7, 14 and 30-day mortality
- Discharge diagnosis confirmation.

To assess the acceptability of the testing process, each participant will be asked the following question:

'How unpleasant was the swab'?

The participants must rate the swabbing process according to the following categories:

1) Very unpleasant

2) Slightly unpleasant

3) I didn’t mind

4) I really didn’t mind.

Thus, the greater the aggregated score for all participants, the more acceptable the swab process was felt to be.

Each participant will also be asked a second question as follows:

‘If you had to have the swab done again, how would you feel about it’?

The participants must give an answer from the following responses:

1) Happy

2) I wouldn’t mind

3) Unhappy.

To assess the acceptability of using the Logicore System, ED research team staff will be asked the following question:

‘How difficult was the Logicore System to use’?

The staff must rate the Logicore System process according to the following categories:

1) Very difficult

2) Slightly difficult

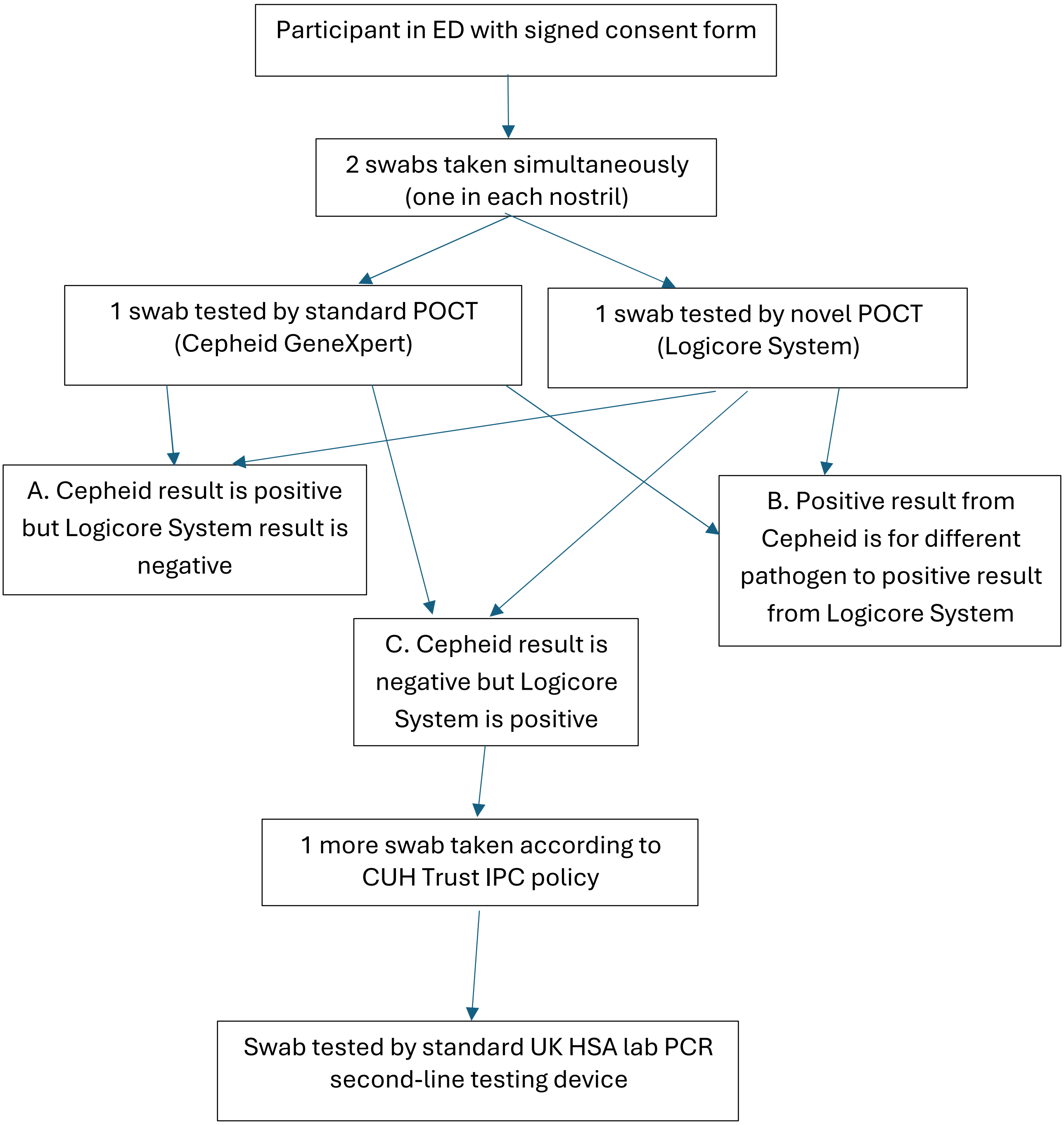
3) Quite easy

4) Very easy.

Thus, the greater the aggregated score for all staff, the more acceptable the Logicore System was felt to be.

The study duration will be a one-off face-to-face visit for participants. The Research Nurse or designated member of the ED research team will use the hospital electronic patient record system to obtain the required post-discharge details. Participants will be considered as in the study from the date of signed informed consent form until the date of discharge from the ED. As there is no additional hospital visit required for the study, there is no provision for participant’s travel or sustenance expenses, and participants will not be paid to take part in the study.

Figure 1: Sample Collection and Testing Process



If Cepheid and Logicore System results are both positive for the same pathogen, or both negative for all pathogens then the results will simply be recorded.

A. If the Cepheid result is positive but the Logicore System result is negative, the Logicore System test will be classed as a false negative.

B. If the Cepheid and Logicore System results are positive for different pathogens, it is not possible to test the participant again as the result from the gold standard Cepheid test will be taken as correct and used to determine appropriate treatment. The participant would not have repeat testing according to standard care. If the Logicore System result is positive for one of the pathogens that Cepheid does not test for (i.e. *Mycoplasma pneumoniae* or human adenovirus), it will be taken to be a true positive result. If however, the Logicore System result is positive for a different pathogen that Cepheid does test for (i.e. Flu A, Flu B, RSV or SARS-CoV-2) but it is not the same pathogen that is positive according to the Cepheid result, the Logicore System result will be classed as a false positive.

C. If the Cepheid result is negative but the Logicore System result is positive, and the participant still exhibits clinical symptoms of respiratory infection requiring diagnostic testing as per CUH Trust Infection Prevention and Control Guideline, a second single swab will be taken from the participant and used for second-line testing as per the CUH Trust standard UK HSA laboratory respiratory virus PCR testing policy. The result from the second-line testing will then be compared to the initial Logicore System result. If the Logicore System result agrees with the second-line result, it will be classed as a true positive. If it does not match the pathogen from the second-line testing, the initial Logicore System result will be classed as a false positive.

The current gold standard POCT at CUH is Cepheid and the second-line PCR test is Luminex, as specified in their IPC Guideline. However, it must be noted that this study is taking place in a real-world setting which may be subject to change due to NHS supply issues, purchasing changes or other factors outside the control of the ED research team. This has been considered during the design of this protocol. If the second-line testing device happens to change during the course of recruiting participants into this study, the replacement second-line device will still need to test for the 2 additional pathogens that the Logicore System tests for. If the second-line testing device changes during the study, the sample collection and testing process shown in Figure 1 will not change. Thus, the Logicore System results will always be compared to the CUH gold standard POCT and when required, to the standard of care second-line testing device that is being used to identify infectious cases in the ED.

12.0 SUBJECT ELIGIBILITY CRITERIA

12.1 Inclusion Criteria

1) Male or female

2) Any age

3) Presenting to CUH Emergency Department

4) Symptomatic of respiratory tract infection as evidenced clinically by the presence of any of the following indicators:

- Acute onset persistent cough (with or without sputum)
- Hoarseness
- Nasal discharge or congestion
- Shortness of breath
- Sore throat
- Wheezing
- Sneezing
- Persistent Acute respiratory distress syndrome
- Influenza like illness
- Fever ≥ 37.8°C
- Any other symptom known to be indicative of acute respiratory episode such as palpitations, headache, anosmia (COVID) or gastrointestinal symptoms (diarrhoea) (influenza)

5) Signed consent form for participation

6) Requiring standard of care diagnostic testing as per CUH Trust IPC policy

7) Able to read and/or understand the age-appropriate participant information sheet in English.

12.2 Exclusion Criteria

1) Unwilling or unable to comply with study nasopharyngeal swabbing procedures

2) Those who are incapacitated or deemed to be lacking capacity to provide informed consent to participate

3) Prisoners or young offenders.

13.0 STUDY PROCEDURES

13.1 Patient Screening and Recruitment

The Investigator, Research Nurse or designated member of the research team will identify potential participants who present at the Emergency Department with suspected acute respiratory tract infection. The potential participant will be screened against the inclusion/exclusion criteria to determine their suitability for recruitment into the study. Potential participants will be provided with a copy of the patient information sheet for the study (as appropriate for their age group) and given as much time as they require to ask any questions and consider whether they want to take part or not. Following the informed consent process and signing of the consent form, the participant will be considered to have been recruited and enrolled into the study and will be allocated a 3-digit study identification number.

13.2 Informed Consent Process

As the study includes participants of all ages, it is vital that an age-appropriate Participant Information Sheet (PIS) is given to the participant and/or parent/legal representative from the following categories:

1) Adults aged 16 years and above

2) Children aged 11 to 15 years of age

3) Children aged 5 to 10 years of age

4) Parent/legal guardian of children 15 years of age or below.

All versions of the PIS and corresponding Informed Consent/Assent Forms must be prospectively approved by the Research Ethics Committee (REC) in line with Good Clinical Practice (GCP), local regulatory and legal requirements. The Investigator or designee must ensure that each study participant, and/or parent/legal guardian if the participant is aged 15 years or less, is fully informed about the nature and objectives of the study and possible risks associated with their participation.

The Investigator or designee will obtain written informed consent from each participant aged 7 years and above, and/or the parent/legal guardian if the participant is aged 15 years or below, before any study-specific activity is performed. The Investigator will retain all original signed consent forms in the Investigator Site File. Each participant will be given a photocopy of their own signed consent form, and a scanned copy will be added to each participant's electronic patient medical record.

If any new information about the Logicore System becomes available during the course of recruitment into the study, which might affect the participant's willingness to continue participating in the study, it will be communicated to the participant and/or their parent/legal guardian as soon as possible. As participants are only involved with the study during one visit to the ED, it is unlikely this information will affect those who have already been recruited into the study. However, if it is deemed appropriate, depending on the nature of the new information, this will be communicated to previous participants verbally via the telephone or by written follow up letter if necessary. Potential new participants will be informed of the new information during the informed consent process, so they are fully appraised of the most up-to-date details. The patient information sheets would be amended accordingly and submitted to the REC for review and approval as quickly as possible.

13.3 Withdrawal of Consent

Participants may withdraw from the study at any time at their own request, (or at the request of the parent/legal guardian if the participant is aged 15 years or less) without providing a reason and without any prejudice as to their further medical care and treatment. Similarly, participants may be withdrawn from the study at any time at the discretion of the Investigator or Sponsor for safety, behavioural or administrative reasons.

If a withdrawn participant agrees, any samples collected as part of this study prior to participant withdrawal will be retained, analysed and used by the study team for the purposes of this study. However, if a participant requests that their research sample is not tested and is destroyed, this will be performed and confirmed by the study team. Participants' samples for standard Cepheid testing will not be destroyed. These samples are needed for standard Trust Infection Prevention and Control testing and will therefore always be tested in line with Trust policy. Participants who withdraw from the study prior to sample collection, or who withdraw after sample collection and request their research sample is destroyed, will be replaced. A new unique study identifying number will be allocated to the replacement participant to prevent any confusion and ensure complete transparency regarding number of participants screened, recruited and withdrawn.

13.4 Randomisation

There are no treatment arms in this study and no randomisation procedure. All participants will be swabbed to provide samples that can be tested by the standard test procedures (Cepheid GeneXpert +/- second-line testing device as per Trust IPC policy) and the novel Logicore System testing device. In this way, each participant will act as their own control for comparison of results.

13.5 Schedule of Events

13.5.1 Per Participant Activities

Activity	At ED Visit
Review of PIS and signing of ICF/Assent form	X
Signs and symptoms of acute respiratory tract infection	X
Temperature	X
Heart rate, respiratory rate, blood pressure and oxygen saturation	X
Nasopharyngeal swabs x2 (simultaneously taken) for initial testing by standard Cepheid device and novel Logicore System device	X
Additional nasopharyngeal swab for testing by second-line device*	X
Assessment of acceptability (Likert-scale)	X
Adverse event details	X

*Additional nasopharyngeal swab may or may not be required, depending on the results from the initial testing and depending on the clinical status of the participant according to Trust IPC policy.

13.5.2 General Study Activities

Activity	Undertaken By	Time Allocation
Informed Consent / Assent*	CI/RN	15 minutes
Study visit procedures and source documentation*	CI/RN	1 hour
Data input into eCRF*	CI/RN	30 minutes
Maintenance of ISF and Essential Documentation^	CI/RN	30 minutes
Query Resolution (following monitoring visit)^	CI/RN	30 minutes

*per enrolled participant.

^per monitoring visit.

13.5.3 Study Sample Collection and Testing Process

Study staff undertaking the sample collection will wear appropriate personal protective equipment (PPE) for the risk exposure at all times, in line with CUH local guidelines for Infection Prevention and Control. To eliminate potential differences in the samples being tested, two nasopharyngeal swabs will be collected simultaneously. One swab will be tested by the gold standard Cepheid multiplex POC test (i.e. standard of care diagnostic testing as required by CUH Trust IPC policy) and the other with the novel Logicore System multiplex POC device. The Logicore System swab will be tested using a specific Logicore System buffer which will be provided for use in a closed vial by the Sponsor. The testing of each specimen (Cepheid or Logicore) must begin within 20 minutes from the time of swab collection, according to the specimen processing guidelines. There is no need to store the swabs/buffer solutions on ice. These can remain at ambient temperature for storage.

The Cepheid POCT is done in the laboratories above the ED. The Logicore System (i.e. the Analytical Module into which the Respiratory Pathogen Panel cartridge is loaded for analysis and the portable Operation Module with display screen, into which the cartridge details and participant anonymised details are input) will be located in a secure clinical area within the ED, with controlled access.

13.5.4 Participant Follow Up

Participants enrolled into the study will be considered to have completed their participation in the study once all the required swabs have been tested and results obtained. (This may simply be one set of 2 swabs, or one set of 2 swabs for initial Logicore System and Cepheid testing plus an additional single swab for second-line testing, if the initial Cepheid result was negative but the participant continues to exhibit clinical symptoms of respiratory infection and needs to be tested as per Trust IPC guideline). Any positive results will be communicated to the participant's medical care team and documented in their electronic medical records. No follow up is required for the study. Participants will continue to receive appropriate medical care outwith the study. This study has no bearing on the clinical management of the participants.

13.5.5 Definition of End of Study

The end of this study is defined as 30 days after the date when the last recruited participant has been discharged from the Emergency Department i.e. the last data point has been collected for the last study participant.

14.0 STATISTICAL ANALYSIS PLAN

14.1 Rationale

According to the Office of National Statistics report on the results of the 2021 Census[7], the population of Cambridge is approximately 146,000 people. Of this population, around 75% classed their ethnicity as White (comprising the following categories: British, Irish, Roma, Gypsy or Irish Traveller, or Other) and around 15% are children aged 15 years or less. For this clinical study, it is expected approximately 20% of participants will be less than 16 years of age, and the majority of participants will be white Caucasian, providing a casemix with external validity in line with the demographics of the catchment area.

This prospective paired diagnostic accuracy and economic evaluation study is designed to assess the clinical performance and potential health system impact of a new molecular Point Of Care Test for respiratory infections in the Emergency Department (ED) setting. The new Logicore System testing device detects six key respiratory pathogens including four infections currently targeted by the current gold standard test (Cepheid); Influenza A (Flu A), Influenza B (Flu B), Respiratory Syncytial Virus (RSV) and SARS-CoV-2. The Logicore System device detects two additional pathogens not included in the established POCT; *Mycoplasma pneumoniae* (MP) and human adenovirus (HAdV).

During the winter season planned for this study, the overall positivity of the current POCT for respiratory disease is estimated at 30-50% of those selected by the respiratory criteria for the standard test (CUH personal communication). However, it is recognised that infections do not distribute evenly by age, or season, or time of the season. For example, RSV is more common in the very young or elderly and has peaks later in the winter months. Thus, the statistical approach to assessment requires us to estimate how many true positives <u>per infection type*</u> might be expected to achieve a sensitivity of 95% with a 10% margin of error, not just overall.

Also, during the winter season, disease type incidence will occur in runs and hence it is possible to inadvertently populate the whole study with one type of infection.

Logically our total sample size will need to be calculated on the lowest prevalence infection. As this is planned as an Intention To Treat study (ITT), i.e. including clinical benefits, then 10-15% extra samples would provide a reasonable margin of samples to allow for withdrawals and invalid samples.

***Table showing impact of variable distribution of infection type on defining sample numbers (after CUH data)**

Infection	Estimated Prevalence in Winter Population	Number Needed for 19 Positives
RSV	~15%	~127
Flu A	~10%	~190
Flu B	~5%	~380
SARS-CoV-2	~10%	~190

As the basis of this study is to determine sensitivity of the Logicore System testing device, rather than combined diagnostic performance across all four infections (rather than each one separately) then we need to apply this distribution imbalance to the way we calculate the minimum sample number to ensure at least achieving our sensitivity goal for each pathogen.

All eligible participants, i.e. those selected by the respiratory and inclusion criteria noted above with respiratory symptoms and a signed consent form, will be enrolled in the study. Using dual swabs, respiratory samples will be collected simultaneously for use in both new (Logicore System) and gold standard (Cepheid) tests to ensure comparability. This method will reduce the risk that differences in test outcomes are the result of inequitable sampling.

The aim also is to use the inclusion/exclusion criteria to recruit consecutive patients in the ED, suspected of the target condition to attempt to reduce spectrum bias. This real-world sample ensures external validity.

As this POCT occurs in a real time ITT setting, sample swabs will be handled consecutively within a few minutes to enter them into the POCT devices with no delay before and between test commencing. Both devices are expected to deliver a result with a similar elapsed time envelope. Hence commonly both tests will be running in parallel at any moment and model more closely the normal pathways of care in use.

This study is primarily aimed at validating the performance of a new POCT device (Logicore System) against an existing POCT device (Cepheid). This is the basis for this statistical plan rather than using a reference standard/composite diagnostic test e.g. PCR + Chest X-Ray.

Blinding the study will be difficult because of the situation of the study.

With regards to incorporation bias, the new test result will not influence the gold standard result.

14.2 Study Endpoints

14.2.1 Specificity (or Sensitivity)

The Primary Endpoint is to demonstrate the sensitivity and diagnostic accuracy of the new Logicore System POCT device compared to the current gold standard (Cepheid) for the four overlapping infections. Based on an expected sensitivity of 95% and a 10% allowable margin, approximately 19 positive cases per infection are required to estimate sensitivity with a 95% confidence. Assuming a positivity rate of up to 50% based on the selection criteria, 400 patients will ensure sufficient numbers of true positive cases for each pathogen and enable accurate estimation of sensitivity, specificity and predictive values for both tests. The paired design allows for direct comparison of the new and established test results using McNemar's test and Cohen's Kappa.

As a secondary component we will evaluate turnaround time, patient and staff acceptability of the new test, and Cost-Benefits of the new test device.

14.2.2 Turnaround Time

Time from swab collection to result availability will be recorded in minutes for both the new POCT device and the established testing device. As both tests are conducted on the same participant, paired comparisons will be analysed using the paired t-test (or Wicoxon's if data are non-normally distributed). Mean (or median) differences will be reported with 95% confidence intervals and p-values.

14.2.3 Acceptability

Participants will complete a brief 'questionnaire' assessing the acceptability of the new POCT device including comfort and willingness to repeat the test. Responses will be collected using Likert scales and categorical items. Data will be analysed using descriptive statistics and comparisons with the standard test will be conducted using Wilcoxon's signed-Rank test using JASP software.

14.2.4 Cost Benefit Analysis

We will evaluate the health economic implications of using the new POCT device to detect two additional pathogens earlier in the patient's care pathway. In the current clinical pathway patients with negative results on the standard test and ongoing symptoms may undergo delayed or reflex laboratory testing to identify other infections, including pathogens targeted by the new test. By enabling earlier diagnosis and treatment at the point of care, the new test may reduce time to treatment, avoid unnecessary empirical therapies and improve resource utilisation. This component will be assessed through a within-trial cost effectiveness analysis using trial data to compare diagnostic pathways and estimate cost per additional case detected, cost per timely treatment initiated and incremental cost-effectiveness ratio (ICER).

The overall hypothesis is that the new test will demonstrate superior sensitivity for RSV based on preliminary data and known limitations of the current test and show the added benefits of the earlier diagnosis on the hospital costs and prescribing.

14.3 Sample Size Calculations

These calculations are based on the expected sensitivity/specificity and desired precision. Methods used are those described by Buderer (1996) and/or Bujang & Adnan (2016) for diagnostic test evaluation.

14.3.1 For Sensitivity (or Specificity)

$$n= (Z^2.P. (1-P))/ d^2$$

n = number of diseased (or non-diseased) subjects required

Z = 1.96 for 95% confidence (the assumption is that this approach could apply to either sensitivity or specificity but here we expect sensitivity will require the higher resolution)

P = expected sensitivity (or specificity) (0.95) for two tailed test

d = desired precision (0.10)

Samples needed for positive cases (sensitivity):

n = 18.2 >>>>19 cases.

From our above table our minimum expected prevalence for any one type of disease is 5% and our maximum is 15% so we can easily estimate what sample total will most likely deliver 19 cases.

If prevalence is low:

N = n/prevalence = 19/.05 = 380 cases.

If the prevalence is uniformly high:

N = n/prevalence = 19/.15 = 127 cases.

This gives us a range of sample size which allows for this variable prevalence. If we assume the worst prevalence state, then 380 cases are required and a further 20 might provide additional safety to achieve a 19-event incidence for each type of infection.

Thus, the recommendation is for a trial recruiting **400** participants.

14.3.2 Turnaround Time

Time from swab collection to result availability will be recorded in minutes for both the new POCT and the established testing device. As both tests are conducted on the same participant, paired comparisons will be analysed using the paired t-test (or Wilcoxon's if data are non-normally distributed). Mean (or median) differences will be reported with 95% confidence intervals and p-value.

14.3.3 Patient Acceptability

Participants will complete a brief questionnaire assessing the acceptability of the new POCT including comfort and willingness to repeat the test. Responses will be collected using Likert scales and categorical items. Data will be analysed using descriptive statistics, and comparisons with the standard test will be conducted using Wilcoxon's signed-Rank test using JASP software.

14.3.4 Cost Effectiveness Analysis

We will be using real patient data on those who fail the original test but are positive on the new test device and have ongoing symptoms requiring further hospital activity and testing. Potentially some may spontaneously improve after the first test and be lost to this analysis.

The data may consist of individual measures of Length Of Stay (LOS), complications, time to treatment etc.

$$n = \left(\frac{(Z_{1-\alpha/2} + Z_{1-\beta}) \cdot \sigma}{\Delta} \right)^2$$

The sample size depends on:

σ = the SD of the effect or costs

Δ = minimally important clinical difference.

For example: cost saving £500 with an SD of £1000 then n = 31.4 or a requirement of 32 per group to analyse.

If we argue that a maximum of 5% of cases will fail the standard test but be positive on the new test and reflex test, then at around 400 patients total we should hope to see 20 - 25 patients per extra infection which is enough to evaluate impact meaningfully.

14.4 Stopping or Early Assessment

Consideration will be given to earlier assessment at say 200 events where there is evidence that the prevalence estimates for each type of pathogen above are out by more than 50%.

14.5 Statistical Methods

14.5.1 Primary Outcomes

The outcome data from each testing device will be compared against its paired standard to produce 2 x 2 tables for each type of disease

	Disease +ve	Disease -ve
	Standard positive	Standard negative
Test positive	True Positive (TP)	False Positive (FP)
Test negative	False Negative (FN)	True Negative (TN)

From this we calculate:

i. Sensitivity, Specificity

Sensitivity is the percentage of cases that had the observed outcome was correctly predicted by the model (i.e., TP/(TP+FN).

Specificity is the percentage of observations that were also correctly predicted as not having the observed outcome (i.e., TN/FP+TN).

ii. Positive Predictive Value

iii. Negative Predictive Value

iv. Likelihood Ratios (LR+ / LR-) for the strength of the test versus standard

v. Accuracy and Cohen Kappa (for agreement)

vi. If data allows, we intend to model some ROC curves around the various clinical thresholds to ascertain the best use model for this device in this clinical setting.

These core calculations and tests will be performed using JASP software on the recorded data sets.

vii. McNemar's test to compare sensitivity/ specificity.

McNemar's test will be used to assess **paired differences in positive and negative results** between the new POCT and the established gold standard test for each of the four shared respiratory infections. This test focuses on cases where the two tests disagree, quantifying whether the new test detects significantly more or fewer positive results than the comparator.

For example, among patients tested for RSV, if the new test identifies more positive cases than the gold standard, and this difference is statistically significant on McNemar's test ($p < 0.05$), this may indicate **superior sensitivity**. Results will be reported with the test statistic, degrees of freedom, and p-value. A similar analysis will be repeated for each infection, supporting the primary endpoint of diagnostic accuracy.

McNemar tests the **null hypothesis** that both tests have the same proportion of positive results by evaluating a test statistic and comparing this with a Chi squared distribution statistic if the value is lower then the null hypothesis is proved.

14.5.2 Turnaround Time

Time from swab collection to result availability will be recorded in minutes for both the new POCT and the established test. As both tests are conducted on the same participant, paired comparisons will be analysed using the paired t-test (or Wilcoxon's if data are non-normally distributed). Mean (or median) differences will be reported with 95% confidence intervals and p-values.

14.5.3 Patient Acceptability

Participants will complete a brief questionnaire assessing the acceptability of the new POCT including comfort and willingness to repeat the test. Responses will be collected using Likert scales and categorical items. Data will be analysed using descriptive statistics and comparisons with the standard test will be conducted using Wilcoxon's signed-Rank test using JASP software.

14.5.4 Cost Benefit Analysis

We plan to evaluate whether detecting the 2 additional infections earlier using the new test device (instead of after standard of care testing in the established pathway brings clinical and economic benefits. Thus, in current practice patients with a negative POCT but remaining symptomatic will normally undergo <u>additional testing</u> (delayed or reflex testing) and this may influence their hospital stay and or ongoing care. With the new testing device these two infections are detected <u>immediately at first presentation</u>.

Our approach with this would be to conduct a cost effectiveness analysis (CEA) or decision-analytic model comparing the effective two pathways.

<u>Current Pathway</u>	<u>New Test pathway</u>
Established test	Single test detects all 6 infections
If Negative: reflex testing	Earlier diagnosis and treatment

Delayed Diagnosis > delayed treatment > possibly worse outcomes.

As this study is really about single timepoint decisions then a simple decision tree would be suitable:

The inputs required:

Test costs: new vs established + reflex testing

Treatment costs

Time to result

Hospital rates (episode and LOS etc)

Complications avoided with earlier treatment

Utility values (QALYS) or similar health outcomes

Probabilities of each infection and outcome.

From this we can calculate:

Incremental Cost-Effectiveness Ratio ICER

Cost per QALY

Budget impact analysis (however if the device behaves as we expect the numbers will be small).

14.6 Estimand Table

Objective	Treatment / Strategy	Population	Variable (Outcome)	Intercurrent Events	Summary Measure (Estimand)
Primary: Compare diagnostic sensitivity and accuracy of new POCT vs gold standard for 4 respiratory infections	New POCT vs gold standard POCT	ED patients with symptoms meeting inclusion/exclusion criteria	Diagnostic test result (positive/negative) per pathogen	- Invalid swab or test - Withdrawal before testing - Gold standard indeterminate	Difference in sensitivity and specificity; 95% CIs for each; McNemar's test for paired comparisons
Secondary 1: Compare turnaround time from swab to result	New POCT vs gold standard POCT	Same as above	Time (minutes) from sample collection to result availability	- Missing timestamps - Result delayed due to user error or system failure	Mean/median time per method; Paired t-test or Wilcoxon signed-rank test
Secondary 2: Compare specificity of new POCT vs gold standard	Same as above	Same as above	Diagnostic result per pathogen in gold standard negative patients	- Invalid test - Patient lost to follow-up	Specificity per infection type; 95% CI
Secondary 3 & 4: Estimate PPV and NPV of new POCT	New POCT	Same as above	PPV = TP/(TP+FP); NPV = TN/(TN+FN)	- Inconclusive reference test result	PPV, NPV per infection; descriptive analysis with 95% CI
Secondary 5: Assess patient acceptability of new test	New POCT	Patients providing questionnaire data	Self-reported scores on comfort, experience, willingness to retest	- Non-response - Incomplete questionnaire	Median scores; frequencies; thematic analysis for free text

15.0 ADVERSE EVENTS AND REPORTING

15.1 Definition of an Adverse Event

As per MHRA Clinical Trials Guidance[8], an Adverse Event (AE) is any untoward medical occurrence, unintended disease or any untoward clinical sign in a clinical investigational subject, temporally associated with the subject's participation in research, whether or not it is considered related to the study procedures, investigational medical device or comparator.

15.2 Definition of a Serious Adverse Event (SAE)

A Serious Adverse Event (SAE) is any untoward medical occurrence that results in any of the following:

1) Death

2) Life-threatening illness or injury (at the time of event; not which hypothetically might have caused death if it were more severe)

3) In-patient hospitalisation or prolongation of existing hospitalisation

4) Permanent impairment of a body structure or body function

5) Congenital abnormality or birth defect

6) Any other important medical event, considered to be serious by the Investigator.

15.3 Definition of an Adverse Device Effect (ADE)

An Adverse Device Effect (ADE) is an Adverse Event (AE) related to use of an investigational medical device (including an IVDD). This includes any AE resulting from insufficiencies or inadequacies in the instructions for the use, deployment, operation or any malfunction of the investigational medical device. This includes any AE that is a result of a user error or intentional misuse of the device.

15.4 Definition of a Serious Adverse Device Effect (SADE)

A Serious Adverse Device Effect (SADE) is an adverse device effect that results in any of the following:

1) Death

2) Life-threatening illness or injury (at the time of event; not which hypothetically might have caused death if it were more severe)

3) In-patient hospitalisation or prolongation of existing hospitalisation

4) Permanent impairment of a body structure or body function

5) Congenital abnormality or birth defect

6) Any other adverse device effect considered to be serious by the Investigator.

As the Logicore POCT device study does not involve an Investigational Medicinal Product, non-serious Adverse Events will not be collected in the study Case Reporting Form (CRF). Only those Serious Adverse Events and Serious Adverse Device Effects (SAEs and SADEs) as defined above, that are suspected to be specifically related to the study procedures or the study device, in the opinion of the Investigator, will be collected in the CRF and reported accordingly.

It is recognised that there may be many deaths in hospital due to infection caused by respiratory pathogens, but the death of a participant in this study would only be relevant if it was deemed due to the procedures or investigative device used for the study. This pragmatic approach to the proportionate reporting of SAEs and SADEs is in line with the updated 'ICH Guideline for Good Clinical Practice E6 (R3)'[9] which has already been implemented in the EU, and which will be adopted in the UK in 2026.

15.5 Definition of Device Deficiency (DD)

Device deficiency (DD) is the inadequacy of a medical device related to its identity, quality, durability, reliability, safety or performance, such as malfunction, misuse or use error and inadequate labelling. All occurrences of Device Deficiencies must be reported to the Sponsor using a DD Reporting Form (blank DD reporting forms will be provided to the site in the ISF). All occurrences of DD should be recorded for study purposes.

15.6 Definition of an Unanticipated Serious Adverse Device Effect (USADE)

An Unanticipated Serious Adverse Device Effect (USADE) is a Serious Adverse Device Effect which by its nature, incidence, severity or outcome has not been identified in the current version of the risk analysis report for the device being tested. All USADEs should be reported to the Sponsor using an SAE/SADE Reporting Form (see section 15.10).

The only expected occurrence relating to the study procedure is discomfort from obtaining the nasopharyngeal swab. In a small number of cases, the participant may experience bleeding from the nostril. However, this will be the same as for the gold standard test procedure and discomfort is likely to only last for a few seconds. Emergency Department staff are well experienced in managing those who may experience nose bleeds.

15.7 Classification of Adverse Event Causality

The Investigator is required to provide their opinion as to the likely causality between the Logicore System IVDD and the SAE/SADE. The classification of the relationship is categorised as follows:

1) Unrelated
2) Unlikely to be related
3) Possibly related
4) Probably related
5) Definitely related.

15.8 Classification of Adverse Event Seriousness

The Investigator is required to classify the seriousness of the SAE/SADE using the CTCAE v5.0 for the most appropriate grading from 1 to 5, where 1 is less serious and 5 is more serious. It should be noted that the term ‘seriousness’ is not the same as ‘severity’. An AE may be considered severe but could be of minor medical significance. An AE is only considered to be serious if it results in any of the points as listed in 15.2, such that an AE can be considered severe but not serious, and vice versa.

15.9 Classification of Adverse Event Severity

The Investigator is required to classify the severity of the SAE/SADE. The severity is related to the intensity of the event, usually classified as mild, moderate or severe. Further guidance as to the classification is as follows:

- Mild: The participant is aware of the event or symptom, but it is easily tolerated
- Moderate: The participant experiences sufficient discomfort to interfere with or reduce their usual level of activity
- Severe: The participant experiences significant impairment of functioning and is unable to carry out their usual activities and/or the participant’s life is at risk.

15.10 Reporting Requirements for SAEs/SADEs

SAE/SADEs (including Unexpected Serious Adverse Device Effects; USADEs) must be reported to the Sponsor if they are deemed to be specifically related to the study procedures or the study investigative device. In such cases, the Investigator or designee must submit an initial SAE/SADE report to the Sponsor as soon as possible (within 24 hours of knowledge of the SAE/SADE taking place). A follow up form should be submitted to update the Sponsor as to further information within 7 days and as more information is available until the conclusion of the SAE/SADE. Blank SAE/SADE Reporting Forms will be provided to the site in the ISF. A new form should be used for each SAE/SADE and when providing follow up details.

The Sponsor must submit an initial report for each SAE/SADE to the MHRA and REC whether or not the Sponsor considers it to be related to the Logicore System IVDD. A follow up report for each incident should be submitted to the MHRA and REC to provide full details of the investigation and the outcome of the SAE/SADE.

The Common Terminology Criteria for Adverse Events (CTCAE) v5.0 will be used to determine the most appropriate medical term for the event, along with the seriousness grading.

16.0 ETHICS AND REGULATORY COMPLIANCE

All performance evaluation studies of an *in vitro* diagnostic device involving participants in an NHS setting require the following approvals to be in place before the study can be declared open to recruitment:

- Research Ethics Committee (REC) approval
- Health Research Authority (HRA) approval
- Local NHS Trust approval: agreement of costings using the National Contract Value Review (NCVR) and signed contract using the model Clinical Trial Agreement (mCTA).

In addition, the MHRA must be informed that the study is taking place.

16.1 Research Ethics Committee and HRA Approvals

The Sponsor will ensure REC/HRA approvals are in place prior to undertaking this clinical study. All correspondence with the REC will be retained in the Sponsor's Trial Master File (TMF) and the site's Investigator Site File (ISF).

16.2 MHRA Notification

Before the study can begin, the Sponsor will inform the MHRA of their intention to perform the clinical study. The MHRA will provide confirmation of receipt but is not required to produce a 'Letter of No Objection' for a performance evaluation of an *in vitro* diagnostic device. Therefore, there is no requirement for regulatory authority to proceed.

16.3 Protocol Amendments

If it becomes necessary to make any substantial changes to the protocol or associated documentation during the study (such as patient information sheets and informed consent forms), the Sponsor will submit a Notice of Substantial Amendment to the REC and local hospital Trust to gain approval for the amendment. All approvals must be obtained before the amendment can be implemented. The MHRA will be informed of the amendment for their information.

The only circumstance in which a substantial amendment may be initiated prior to the necessary approvals is when the change is necessary to eliminate an immediate risk to the participants (i.e. an Urgent Safety Measure). In this case, accrual of any new participants will be halted until all required approvals have been obtained.

If the amendment is not classified as substantial (i.e. it is non-substantial), the Sponsor will inform the REC, MHRA and local hospital Trust of the details for their records, but the formal approval process is not required.

Details of all substantial amendments will be immediately included in section 4.0 of this protocol accordingly. Details of non-substantial amendments will be added to section 4.0 if/when the next substantial amendment is required.

16.4 Study Documentation and Reporting

Copies of all correspondence with the REC and MHRA will be kept in the Investigator Site File (ISF) as well as the Trial Master File (TMF). The Chief Investigator will be responsible for submitting a Progress Report to the REC within 30 days of the study being opened. Furthermore, the Chief Investigator will submit an Annual Progress Report (APR) to the REC each year and an End of Study Report within a year of the study being completed. If the study is terminated prematurely for any reason, the Chief Investigator will inform the REC.

16.5 Data Protection

All site staff must comply with the requirements of the General Data Protection Regulation (GDPR), the Data Protection Act 2018 and Trust Policy with regards to the collection, storage, processing, transfer and disclosure of personal information and will uphold the core principles including collecting the minimum data to achieve the aims of the study, ensuring participants understand how their data will be used and maintaining accurate and secure records.

16.6 Participant Confidentiality

When a participant signs the consent form and is enrolled into the study, they will be assigned a unique 3-digit study identifying number, serving to de-identify and pseudonymise the individual. This number will be the only means by which the Sponsor can identifying the participants. The Sponsor will have no access to participant identifiable details.

The site will maintain a subject log, detailing individual participants and their corresponding study identifying numbers. The subject log will not be available to the Sponsor. However, the study monitor will need access to the subject log and the original signed consent forms during the monitoring process to ensure they are reviewing the correct participant's medical records for source data verification purposes. This will be explained to the participants during the consent process.

The subject log containing the link between individual participants and study identifying number will be securely stored in a separate location to the study data, in a password protected digital file. The log will only be available to members of the site study team and the study monitor. Thus, the site is the data custodian for any participant identifiable data and the Sponsor is the data custodian for all de-identified pseudonymised data for the purposes of the study.

Despite employing best practice to minimise the possibility of a breach of participant confidentiality, in the event of identifiable details being passed to anyone outwith the immediate members of the site ED research team, the person discovering the breach must inform the Sponsor of the details within 24 hours. The Sponsor must then determine whether the breach is a high risk to the rights and freedom of the individual/s involved and if so, inform the Information Commissioner's Office (ICO) within 72 hours of the breach being discovered. The individual/s must also be informed of the breach without undue delay. The Sponsor will maintain a log of all participant identifiable data breaches (both high and low risk) that occur during the study.

16.7 Patient and Public Involvement

There was no patient and public involvement in the course of developing the Logicore System *in vitro* diagnostic device for CE marking. It was felt that patients and the public became well versed with PCR testing during the COVID pandemic and understand the basic premise of nasal swabbing to detect pathogens. This process has become widely accepted by the general public.

Prior to undertaking this study however, a snapshot of public opinion was obtained from 22 patients in the ED at Addenbrooke's Hospital. The patients were asked 'If we had 2 devices that could test for a bug like COVID or Flu, but we didn't know which device was the best, would you let us take 2 nasal swabs instead of 1 so we could work out which is best?' The patients were also told the swabs would be taken simultaneously, one from each nostril. 91% (20 out of 22) of patients said 'Yes', they would let us take 2 swabs to work out which device was best. One patient said 'No' and one patient said 'Don't know'. This demonstrates a high level of support, albeit in a small sample of the population, for our study to be performed.

Furthermore, one of the aims of this study is to assess the acceptability of the new device to the participants and staff.

16.8 Peer Review

During the development of this study protocol, the Sponsor consulted with several Consultant Clinicians who are independent of Logilet (UK) Ltd, to ensure the study is designed to assess potential clinical benefit for patients and health economic benefit to the hospital NHS Trust. As there is no Investigational Medicinal Product or other therapeutic agent involved in the study, and little risk to study participants, a formal Data Monitoring Committee or Data Advisory Committee will not be convened. The Sponsor will review study data as it is accrued and will liaise with the site study team if there are any concerns about safety to determine whether it is necessary to halt the study prematurely.

16.9 Audits and Inspections

16.9.1 Protocol Compliance

Protocol deviations, non-compliances, or breaches are departures from the approved protocol. The ISF and participants' source documentation/medical records will be reviewed by the study monitor to assess protocol compliance and ensure the validity of the study data. If the monitor detects any deviation from the protocol, it will be brought to the attention of, and discussed with, the Chief Investigator to determine the reason for the deviation and to mitigate against further occurrences. If possible, the deviation will be corrected. If it is not possible to correct the deviation, the Sponsor will decide whether the data is still suitable for inclusion or should be excluded from analysis.

Similarly, if a member of the site team realises an accidental protocol deviation has occurred, they should immediately notify the Chief Investigator and the Sponsor, providing details of what has happened and how it differs from the approved protocol. The Chief Investigator and Sponsor will discuss whether the deviation can be rectified in any way. If it is not possible to correct the error, the Sponsor will decide whether the data is still useful or whether it should be excluded from the analysis.

The study monitor will provide protocol re-training to relevant team members following a deviation as required. If a member of the study team continues to deviate from the requirements of the protocol despite attempts by the Sponsor to re-train the individual, the Sponsor may request a replacement person to deal with further study participants.

Each Protocol Deviation (PD) will be classed as minor or major by the Sponsor; a deviation will be considered major if the integrity of the study data is deemed to have been jeopardised in any way. Details of major deviations will be reported to the Research Ethics Committee.

In addition, any breaches of Good Clinical Practice (GCP) must be documented and brought to the attention of the Chief Investigator and Sponsor immediately (within 24 hours of detection).

Prospective, planned deviations or waivers to the protocol are not allowed under the UK clinical trial regulations and must not be used.

16.9.2 External Audit

In the event of request for external site audit or formal inspection by any Body, the Trial Master File containing all Essential Documentation (and/or Investigator Site File as required) will be made available for review along with the medical records/source documents for the study participants. Participants will be informed of this possibility during the informed consent process.

16.10 Publication and Dissemination Policy

Ownership of the raw data collected for this study will reside with the Sponsor. On completion of the study the Sponsor will analyse the data and produce a Final Study Report within one year of the end of the study. Results will not be available on an individual participant level. Overall study findings will be made publicly available by publication in peer reviewed journals where possible. In addition, results may be presented at national and international scientific meetings. All publications will acknowledge the vital support of CUH ED Research Team.

Participants that wish to be informed of the results of the study will be given a lay summary of results when they are available, post-analysis. The Sponsor will be responsible for producing the lay summary of results and the site study team will be responsible for providing the summary to individual study participants accordingly.

17.0 INDEMNITY

The Sponsor has obtained an appropriate insurance policy to cover this specific clinical study. The Insurance Certificate will be made available to the Research Ethics Committee and the CUH Trust prior to the commencement of the study at the hospital site. This provides insurance cover for all study participants to ensure the Sponsor is able to pay compensation in the event of a participant experiencing a serious injury as a direct result of taking part in the study.

18.0 DATA COLLECTION, HANDLING AND VERIFICATION

18.1 Source Documentation and Data Verification

Prior to commencement of the study, the site will confirm the location of all original data required by the protocol (i.e. where to find specific source documentation). All source documents must be kept securely and available for review by the study monitor upon request. Fully anonymised data will be transferred into an electronic Case Report Form (eCRF). All study data in the eCRF must be extracted from and consistent with the relevant source documents. The eCRF must be completed by the designated member of the study team in a timely manner (i.e. within one week of date of assessments taking place).

During a monitoring visit, if the monitor detects any discrepancies between the source documentation and data input into the eCRF data, it will be brought to the attention of the relevant study team member/s as a query for resolution. The team will resolve the query within 5 working days of receipt of the query and ensure source documentation and eCRF is updated with the correct information.

A monitoring plan will be generated prior to study commencement by the Sponsor, detailing the frequency and scope of the monitoring for the study. The frequency of monitoring visits by the Sponsor's representative will be determined by an initial risk assessment performed prior to the start of the study. Throughout the course of the study, the risk assessment will be reviewed and the monitoring frequency adjusted as necessary.

18.2 Clinical Site Training

Although not mandatory for studies that do not include an Investigational Medicinal Product, it is recommended that all study staff should hold evidence of appropriate GCP training prior to undertaking any study procedures. Additionally, the Sponsor will perform a Site Initiation Visit (SIV) prior to opening the study, during which the site team will be trained in the study-specific procedures of the protocol. The SIV provides the team with opportunity to ask any questions to ensure understanding of all study requirements. Each member of the study team is required to sign the delegation of duties log and provide evidence of suitability to perform these duties (e.g. current CV and GCP certification). A list of all documentation that is required prior to site opening to recruitment of participants is provided in Appendix 4. The Sponsor will issue a formal letter of confirmation of site authorisation to open the study, after receipt of which the site can commence recruitment of participants.

18.3 Retention of Source Documentation

In accordance with UK legal requirements, all clinical trial documentation and data must be kept for 25 years following completion of the study. The Sponsor and site will agree an archiving process during the Clinical Trial Agreement negotiations, prior to commencing the study at the site.

19.0 REFERENCES

1. The Global Burden of Disease 2004 Update. World Health Organization. ISBN 978 92 4 156371 0.

2. Cambridge University Hospitals NHS Foundation Trust Infection Prevention and Control Guideline: Patient placement guide for respiratory viruses. Version 5; Approved June 2024.

3. GeneXpert Xpress SARS-CoV-2/Flu/RSV Instructions For Use. Cepheid Rev. A October 2020.

4. REGULATION (EU) 2017/746 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 5 April 2017 on *in vitro* diagnostic medical devices and repealing Directive 98/79/EC and Commission Decision 2010/227/EU.

5. World Medical Association Declaration of Helsinki – Ethical Principles for Medical Research Involving Human Participants 2024.

6. International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) – ICH Harmonised Guideline – Integrated Addendum to ICH E6(R1): Guideline for Good Clinical Practice E6(R2) Current *Step 4* version dated 9 November 2016.

7. Office for National Statistics – How life has changed in Cambridge: Census 2021 https://www.ons.gov.uk/visualisations/censusareachanges/E07000008/

8. MHRA Guidance: Clinical trials for medicines: collection, verification, & reporting of safety events. Published 25 June 2025.

9. ICH Harmonised Guideline for Good Clinical Practice E6 (R3) Final Version Adopted on 06 January 2025.

20.0 APPENDICES

20.1 Appendix 1 – Details of Equipment Used in the Study

The gold standard *in vitro* diagnostic device used at CUH, is Cepheid POCT (GeneXpert).

Following a negative Cepheid result, if a patient still exhibits clinical symptoms of respiratory infection, the standard second-line testing device is Luminex NxTag, as per CUH Trust Infection Prevention and Control Guideline at the time of devising this protocol.

The IVDD that is the subject of this investigation is the Logicore System.

The Logicore POCT System will always be assessed against the gold standard first-line POCT and standard of care second-line testing devices in line with the Trust IPC policy.

20.2 Appendix 2 – Logicore System Specimen Processing Instructions

The swab to be tested by the Logicore System device must be processed using the Logicore System buffer solution provided by Logilet. Each sample requires a new Respiratory Pathogen Panel cartridge for the testing and analysis process. Initially it will be possible to run the analysis on a maximum of 2 cartridges at the same time as 2 modules are being provided for the study in the first instance. Once the staff are accustomed to the processes involved, and providing there is sufficient physical space to house the modules, the site may be provided with an extra 2 modules to allow 4 cartridges to be analysed simultaneously. Instructions for Use will be provided to the site staff along with training to ensure the equipment is utilised correctly.

20.3 Appendix 3 – Common Terminology Criteria for Reporting Adverse Events v5.0

This document will be provided to the clinical site as an attachment in electronic form.

20.4 Appendix 4 – Site Authorisation Documentation List

The following documentation must be provided by the site before formal Site Authorisation can be issued to commence the clinical study:

- Protocol Signature Page completed by CI
- CV and GCP certificate for each member of the research team
- Age-appropriate Patient Information Sheets on local headed paper
- Informed Consent/Assent Forms on local headed paper
- Delegation of Authority Log / Site Signature Sheet
- Site Training Log
- Signed model Clinical Trial Agreement (mCTA) including costings as determined by National Contract Value Review (NCVR)
- Local Equipment Validation Documentation.