Protocol

The efficacy and safety of very low dose dexamethasone used to facilitate the extubation of ventilator dependent preterm babies who are at high risk of bronchopulmonary dysplasia.

Sponsor Name: University of Liverpool

Trial Identifiers:

Sponsor Number: UoL001206

EudraCT Number: 2015-005342-63

ISRCTN: ISRCTN81191607

REC Ref: 16/NW/0396

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Abbreviations

Abbreviation	Description
AE	Adverse Event
BAL	Bronchoalveolar Lavage
BPD	Bronchopulmonary Dysplasia
CI	Chief Investigator
CIG	Co-Investigator Group
CP	Cerebral Palsy
CPAP	Continuous Positive Airway Pressure
CRF	Case Report Form
CrUSS	Cranial Ultrasound Scan
DMC	Data Monitoring Committee
DSUR	Development Safety Update Report
GCP	Good Clinical Practice
HR	Hazard Ratio
IL	Interleukin
IMP	Investigational Medicinal Product
IRAS	Integrated Research Application System
IVH	Intraventricular Haemorrhage
MHRA	Medicines and Healthcare products Regulatory Agency
NEC	Necrotising Enterocolitis
NHS	National Health Service
NIHR	National Institute for Health Research
NPEU CTU	National Perinatal Epidemiology Unit Clinical Trials Unit
PDA	Patent Ductus Arteriosus
PEEP	Positive End Expiratory Pressure
PI	Principal Investigator
PIL	Parent Information Leaflet
PMA	Postmenstrual Age
PMG	Project Management Group
R&D	NHS Trust Research and Development Department
RCT	Randomised Controlled Trial
REC	Research Ethics Committee

RR	Risk ratio
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SD	Standard deviation
SUSAR	Suspected Unexpected Serious Adverse Reaction
TNF	Tumour Necrosis Factor
TSC	Trial Steering Committee

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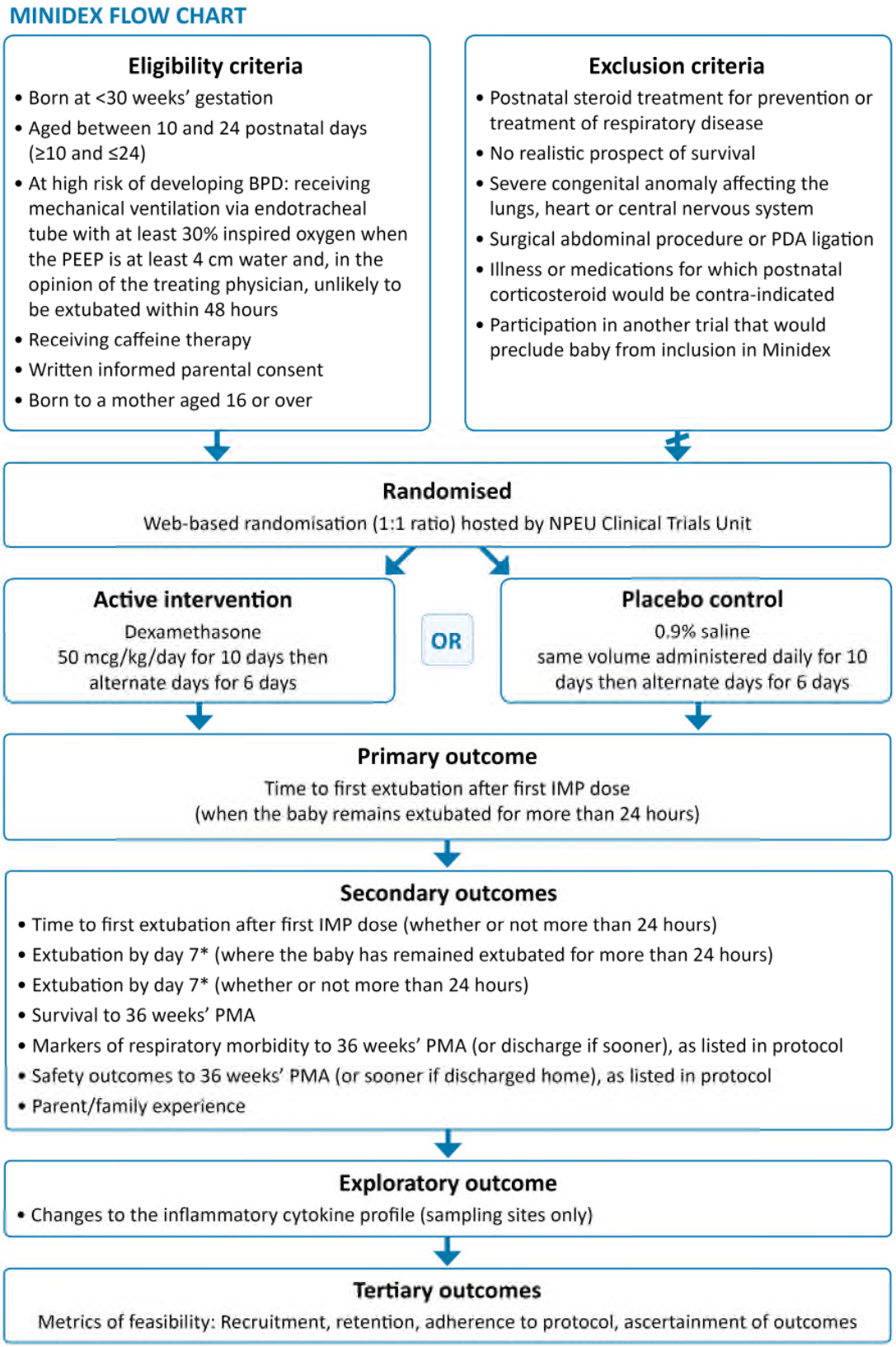
Trial Flow Diagram



*All references to 'day 7' within this protocol refer to 7 completed days from first IMP dose.

Study Summary

GENERAL INFORMATION	
Short Title	Minidex
Full Title	Minidex: The efficacy and safety of very low dose dexamethasone used to facilitate the extubation of ventilator dependent preterm babies who are at high risk of bronchopulmonary dysplasia.
Sponsor	University of Liverpool
Sponsor ID	UoL001206
EudraCT No.	2015-005342-63
REC Reference No.	16/NW/0396
Chief Investigator	Mark Turner
Lead Clinician	Helen Yates
Co-ordinating Centre	National Perinatal Epidemiology Unit Clinical Trials Unit
National / International	National
TRIAL INFORMATION	
Phase	Randomised Phase 2b
Indication	Prevention of bronchopulmonary dysplasia (BPD).
Design	Multicentre, randomised, masked, parallel group, placebo controlled trial.
Primary Objective	To determine if treatment with very low dose dexamethasone facilitates the extubation (for more than 24 hours) of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD.
Secondary Objectives	To determine if the treatment of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD with very low dose dexamethasone impacts upon: 1. Time to first extubation after first IMP dose (whether or not more than 24 hours) 2. Extubation (for more than 24 hours) by day 7 3. Extubation (whether or not more than 24 hours) by day 7 4. Survival to 36 weeks' postmenstrual age (PMA) 5. Respiratory morbidity to 36 weeks' PMA (or discharge home if sooner) 6. Safety outcomes 7. Parent/family experience
Exploratory Objective	To determine if the treatment of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD with very low dose dexamethasone impacts upon cytokine profile.
TRIAL TIMELINES	
Expected start date	01/11/2015

Participant enrolment phase	01/09/2016 – 28/02/2018
Follow-up duration	36 weeks' PMA
End of Trial Definition	Date of final data lock
Expected completion date	30/10/2018
TRIAL PARTICIPANT INFORMATION	
Number of trial participants	94
Number of trial sites	Up to 25
Age group of trial participants	Preterm babies born less than 30 weeks' gestation and aged between 10 and 24 postnatal days.
Inclusion criteria	Babies: 1. Born at <30 weeks' gestation 2. Aged between 10 and 24 postnatal days (≥10 and ≤24) 3. At high risk of developing BPD: receiving mechanical ventilation via endotracheal tube with at least 30% inspired oxygen when the positive end expiratory pressure (PEEP) is at least 4 cm water and, in the opinion of the treating physician, unlikely to be extubated within 48 hours 4. Receiving caffeine therapy 5. Written informed parental consent 6. Born to a mother aged 16 or over
Exclusion criteria	1. Postnatal steroid treatment for prevention or treatment of respiratory disease 2. No realistic prospect of survival 3. Severe congenital anomaly affecting the lungs, heart or central nervous system 4. Surgical abdominal procedure or PDA ligation 5. Illness or medication for which postnatal corticosteroid would be contra-indicated (e.g. active fungal infection, confirmed or suspected acute sepsis, acute necrotising enterocolitis (NEC)/focal intestinal perforation or cyclooxygenase therapy) 6. Participation in another trial that would preclude baby from inclusion in Minidex
INVESTIGATIONAL MEDICINAL PRODUCT	
IMP name(s)	Dexamethasone Saline
Duration of IMP Treatment	16 days 50 mcg/kg administered once daily for 10 days then alternate days for 6 days. Dose to be calculated on current working weight at trial entry; can be increased in line with current working weight as per local practice.
IMP supplier	QP released from Guy's and St Thomas' (manufacturer)
Non IMP name	Caffeine therapy

1. Introduction

1.1. Background

Bronchopulmonary dysplasia (BPD) affects approximately 35% of babies born at less than 30 weeks' gestation who survive to discharge from neonatal care.[1,2] In the UK this equates to 1,450 affected babies a year. BPD prolongs hospitalisation and often requires home oxygen therapy for 6–12 months.[3] It is associated with a significant long term respiratory and neurodevelopmental healthcare burden: each year up to 200 UK babies develop moderate/severe disability attributable to BPD.[4] Therapies that reduce the burden of BPD will reduce the burden of neurodevelopmental and respiratory impairment that it entails. There are few interventions proven to reduce the incidence or severity of BPD.

Corticosteroids have been used in high dose regimens since the 1970s to reduce the impact of BPD. They have been shown in meta-analyses to facilitate the extubation of preterm babies and reduce the incidence of BPD at the expense of short-term metabolic and gastrointestinal side effects. A Cochrane review[5] reported a risk ratio (RR), compared to placebo, for 'failure to extubate by the 7th day' of 0.64 (95% Confidence Interval (CI) 0.56 to 0.74) and a RR for 'reduction in the incidence of BPD' of 0.76 (95% CI 0.66 to 0.88). Facilitation of extubation and reduction of BPD is seen for both hydrocortisone and dexamethasone (at all published doses of dexamethasone, ranging from low (0.15 mg/kg/day) to high (0.6 mg/kg/day)) when administered at any postnatal age[5,6,7]. Corticosteroid therapy became commonplace; in 1995, 72.5% of all surviving UK babies less than 26 weeks' gestation had received postnatal corticosteroids.[8]

It is assumed that the effect of corticosteroids is mediated by amelioration of the lungs' inflammatory responses.[9] BPD is known to be associated with elevated levels of proinflammatory cytokines[10,11] and corticosteroid administration has been proven to reduce levels of these cytokines (e.g. interleukin (IL)-1β, IL-1ra, IL-6, IL-8 and tumour necrosis factor (TNF)α) in affected babies.[12] However, little is known about the cytokine:cytokine network interactions underpinning BPD, and the way in which these are modulated by corticosteroids has yet to be elucidated.

The risk-benefit balance of corticosteroids is complicated by its association with adverse neurodevelopmental outcomes. This was first reported in 1974[13] and in 2001 a meta-analysis[14] reported that babies treated with high dose regimens of postnatal corticosteroids have an increased risk of long-term adverse neurodevelopmental outcomes (Cerebral Palsy (CP), RR 1.92 (95% CI 1.41 to 2.61)). However, further meta-analyses have shown that the risk of adverse neurodevelopmental outcomes is modified by both the postnatal age at which the corticosteroids are given[5,6] and the baby's underlying risk of developing BPD.[15] So the increased risk of adverse outcomes seen in the initial meta-analysis was attributable to those trials that included babies less than 7 days of age, treated to prevent BPD (CP RR 1.75 (95% CI 1.20 to 2.55)).[6] The risk of CP is not increased in those trials treating babies over 7 days of age for evolving BPD (CP RR 1.06 (95% CI 0.76 to 1.5))[5] and the risk of the composite outcome of CP and death was reduced by corticosteroid administration in trials where the background risk of BPD exceeded 65%.[15] Some clinicians advocate giving corticosteroids from birth in order to prevent BPD. Recent data suggests this approach may reduce the incidence of BPD[7]. However, many clinicians are concerned that giving corticosteroids to all babies born at extreme prematurity may expose many babies to corticosteroids unnecessarily. Accordingly, many clinicians use corticosteroids selectively, that is in babies more than 7 days old who are at high risk of developing BPD. Evidence about the efficacy of corticosteroids when administered to all premature neonates is not relevant to corticosteroids given 2 – 3 weeks after birth because responses to corticosteroids will change with time and doses given during the first

week will have effects on adaptation to extrauterine life that are not relevant if steroids are used selectively.

Some clinicians now use low or very low dose regimens to improve pulmonary function in babies at high risk of BPD after day 7 of life in the hope that, like other side effects, the effect on the brain is dose-dependent and low and very low dose regimens will avoid neurodevelopmental impairment while maintaining the beneficial pulmonary effects. There is insufficient evidence to support this. No low dose regimen starting after 7 days has been subjected to a randomised controlled trial adequately powered on neurodevelopmental outcomes. Such a trial is needed to inform clinical practice and this study is the necessary first step towards designing a large trial based on clinically meaningful outcomes.

In a retrospective cohort study we found that babies at risk of BPD who received a very low dose regimen were extubated significantly faster than controls who did not receive corticosteroids, hazard ratio (HR) 6.24 (95% CI 2.34 to 16.63).[16] This regimen, Minidex (50 mcg/kg/day for 10 days then alternate days for 6 days) was derived from the physiological corticosteroid replacement dose, and is lower than the next lowest dose regimen that has been subjected to assessment of short-term outcomes (DART 150 mcg/kg/day with 10 day taper).[17] Our cohort study suggested proof of concept for an effect of a very low dose of dexamethasone on short-term outcomes relevant to BPD. This now needs to be evaluated in a rigorously designed randomised controlled trial.

If the efficacy and acceptability of very low dose dexamethasone (Minidex) is established in this trial, the next step would be to conduct a large pragmatic multicentre randomised placebo controlled effectiveness trial examining substantive outcomes such as neurodevelopment at 2 years of age corrected for prematurity, building on the conduct and findings of this trial.

1.2. Rationale for the Study

Systematic reviews indicate that there is a net benefit of giving postnatal corticosteroids to a group of selected preterm babies at high risk of BPD.[5,15] However, there is a need to determine the optimal dose for postnatal corticosteroids. “High” doses are effective but may be associated with avoidable adverse effects. We will investigate a dosage regimen (Minidex) that is substantially lower than the lowest dose regimen that has been assessed to date. Proof of concept for the effect of Minidex on an appropriate surrogate outcome has been established in a retrospective cohort study.[16] However, there is no definitive evidence of efficacy, size of effect or safety. Our long-term goal is to conduct an effectiveness trial aimed at establishing the 2-year neurodevelopmental outcomes of babies receiving Minidex.

Prior to the effectiveness trial we need to perform an efficacy study to determine short-term clinical efficacy and safety, acceptability to parents and clinicians and feasibility in terms of recruitment and retention. In this efficacy study, extubation and respiratory morbidity prior to hospital discharge are being employed as surrogate markers of pulmonary function. Collection of data regarding respiratory morbidity and the need for open-label medication to hospital discharge will increase our understanding of the biological processes and enable planning of the next step in evaluating this therapy.

This study offers an opportunity to gain a better insight into the inflammatory perturbations found in BPD as well as highlighting the mechanisms of action through which dexamethasone can influence the structure and interactions within inflammatory networks. Characterising key nodal (cytokine) perturbations within these networks among babies treated with dexamethasone or placebo may highlight novel potential targets for therapeutic intervention (i.e. the presence of hub/driver nodes),

or differences in inflammatory signatures between therapy responders versus non-responders. These results may indicate ways therapy could be targeted, avoiding unnecessary exposure in the future.

The aim of the study is to assess the efficacy and safety of very low dose dexamethasone used to facilitate the extubation of ventilator dependent preterm babies less than 30 weeks' gestation at high risk of developing BPD.

2. Trial Objectives

Objectives	Outcome Measures	Timepoint(s) of evaluation of this outcome measure (if applicable)
Primary Objective		
To determine if treatment with very low dose dexamethasone facilitates the extubation (for more than 24 hours) of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD.	Time to first extubation after first IMP dose when the baby remains extubated for more than 24 hours.	
Secondary Objectives		
To determine if the treatment of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD with very low dose dexamethasone impacts upon:		
1. Time to first extubation (whether or not more than 24 hours)	Time to first extubation after first IMP dose (whether or not more than 24 hours)	
2. Extubation by day 7	(a) Extubation by day 7 (where the baby has remained extubated for more than 24 hours) (b) Extubation by day 7 (whether or not more than 24 hours)	Day 7
3. Survival to 36 weeks' postmenstrual age	Alive at 36 weeks' PMA (or discharge home if sooner)	36 weeks' PMA (or discharge home if sooner)
4. Respiratory morbidity to 36 weeks' PMA	(a) Total duration of invasive ventilation	36 weeks' PMA (or discharge home if sooner)

Objectives	Outcome Measures	Timepoint(s) of evaluation of this outcome measure (if applicable)
(or discharge home if sooner)	through endotracheal tube (b) Total duration of non-invasive respiratory support through nasal CPAP, nasal ventilation or high flow oxygen therapy (c) Open-label treatment using local unit policy i.e. escalation of corticosteroid therapy or requirement for repeated courses of corticosteroid. (d) Use of diuretics using local unit policy (e) Total duration of supplemental oxygen therapy (f) Severity of BPD at 36 weeks' PMA (or discharge home if sooner) as defined in Appendix 1. Oxygen requirement will be determined using the oxygen reduction test (see Appendix 2).	
5. Safety outcomes	a.) Hypertension b.) Hyperglycaemia c.) Confirmed/suspected sepsis d.) Spontaneous gastrointestinal perforation or NEC (definitive and/or complicated (Bell stage II and above) confirmed by radiography and/or histopathology or requiring surgery) e.) Deterioration in cranial ultrasound findings. A new finding of severe IVH (grade III/IV with ventricular dilation or intraparenchymal	During administration of IMP and for 14 days after last dose of intervention (a-d) CrUSS performed prior to randomisation (or when occurring according to standard clinical

Objectives	Outcome Measures	Timepoint(s) of evaluation of this outcome measure (if applicable)
	bleeding) or new PVL on the CrUSS performed at 36 weeks' PMA (+/- 2 weeks) when compared to the scan performed most recently prior to randomisation f.) Growth i) Change in standard deviation score (SDS) for weight between study entry and day 14 after randomisation. ii) Change in standard deviation score (SDS) for weight between day 14 after randomisation and 36 weeks' PMA (or discharge home if sooner) ii) Change in standard deviation score (SDS) for head circumference (as measured by occipito-frontal circumference) between study entry and 36 weeks' PMA (or discharge home if sooner)	practice) and at 36 weeks' PMA (+/- 2 weeks, or discharge home if sooner). Weight measured at trial entry, on day 14 (after randomisation) and 36 weeks' PMA (+/- 2 weeks, or discharge home if sooner) Head circumference measured at trial entry, and at 36 weeks' PMA (+/- 2 weeks, or discharge home if sooner)
6. Parent/family experience	Parent/family experience: Measures of the parents'/family's hands on involvement in their baby's care. (a) Duration, frequency and amount of contact with their baby (b) Degree of parental/family involvement in cares (nappy changes, skin care, etc.)	Log recorded during 16-day intervention period. Although part of the consent process, this is not part of the source dataset and is optional for parents and staff to complete

Objectives	Outcome Measures	Timepoint(s) of evaluation of this outcome measure (if applicable)
Exploratory Objective		
Cytokine profile (Consenting babies from specified sites only)	Changes to the inflammatory cytokine profile in blood and endotracheal tube secretion fluid (a mechanistic marker of response)	Endotracheal tube secretions and blood samples taken at trial entry, and on days 4, 7, 10, 14 of the IMP administration schedule
Tertiary Objectives		
To assess the acceptability and feasibility of the trial in order to inform the design of the subsequent effectiveness trial by reporting metrics related to:		
1. Recruitment	Recruitment rate	Trial completion
2. Retention	Retention rate	Trial completion
3. Adherence to protocol	Use of open-label therapy and completion of course	Trial completion
4. Completeness of data	Ascertainment of outcomes	Trial completion

3. Trial Design

This is a multicentre, randomised, masked, parallel group, placebo controlled trial to examine the efficacy of very low dose dexamethasone in facilitating the extubation of ventilator dependent preterm babies.

The trial is anticipated to take 30 months to complete and aims to recruit a total of 94 babies in up to 25 sites across the UK over 18 months.

Parents of babies recruited at specified sites will be asked to consent to their baby having samples taken for cytokine estimation. This will allow modelling of their inflammatory networks.

3.1. Inclusion Criteria

Eligible babies:

1. Born at <30 weeks' gestation
2. Aged between 10 and 24 postnatal days (≥10 and ≤24)
3. At high risk of developing BPD: receiving mechanical ventilation via endotracheal tube with at least 30% inspired oxygen when the PEEP is at least 4 cm water and, in the opinion of the treating physician, unlikely to be extubated within 48 hours
4. Receiving caffeine therapy
5. Written informed parental consent
6. Born to a mother aged 16 or over

3.2. Exclusion Criteria

Ineligible babies:

1. Postnatal steroid treatment for prevention or treatment of respiratory disease
2. No realistic prospect of survival
3. Severe congenital anomaly affecting the lungs, heart or central nervous system
4. Surgical abdominal procedure or PDA ligation
5. Illness or medication for which postnatal corticosteroid would be contra-indicated (e.g. active fungal infection, confirmed or suspected acute sepsis, acute NEC/focal intestinal perforation or cyclooxygenase therapy)
6. Participation in another trial that would preclude baby from inclusion in Minidex

3.3. Setting

The trial will take place in up to 25 tertiary neonatal units across the UK.

4. Trial procedures

4.1. Trial Schedule

Procedure	Pre-randomisation	Post-consent pre-IMP	Days after Randomisation																	36 weeks PMA
			1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17-30	
Eligibility assessment																				
Consent																				
Randomisation																				
Baseline clinical data																				
IMP administration																				
Clinical data collection																				
Measurement of occipito-frontal circumference																				
Measurement of weight																				
Blood sample & ET secretions collection[1]																				
Oxygen reduction test																				
Safety reporting																				

[1]Babies recruited at specified sites only. Samples to be taken within the 4-hour period prior to the IMP dose.

4.2. Recruitment

Daily review of the baby's status will allow the attending clinical team to identify babies likely to be eligible for the trial. Trial eligibility will be confirmed by a medically qualified clinician who is on the delegation log and documented in the baby's Case Report Form (CRF) and medical notes.

Once a baby has been identified as eligible, a full verbal and written explanation of the trial (via the Parent Information Leaflet) will be provided for the parents to consider. Parents who do not speak English will only be approached if an unrelated adult interpreter is available.

4.3. Consent

Following provision of information about the trial, parents will have time to consider participation and will be given the opportunity to discuss the trial with their family and healthcare professionals before they are asked whether they would be willing to take part in the trial.

Parents of eligible babies who are interested in the trial will provide informed, written consent by means of a dated signature. The right of the parents to decline participation without having to give a reason will be respected. For those babies who are randomised, their parents will remain free to withdraw them from the trial at any time without giving reasons and without prejudicing any further treatment.

The written consent will be taken by the Principal Investigator or appropriately qualified healthcare professional who has delegated authority. The process of obtaining written consent will be clearly documented in the baby's medical notes. One copy of the completed consent form will be given to the parents, one filed in the Site File, one filed in the hospital notes and the original signed copy sent to the National Perinatal Epidemiology Unit Clinical Trials Unit (NPEU CTU) in Oxford for the Trial Master File.

4.4. Randomisation

Babies will be randomised in the ratio 1:1 to receive either very low dose dexamethasone or a matched placebo. Allocation will be masked from the baby's family.

Randomisation will be managed via a secure web-based randomisation facility hosted by the NPEU CTU, University of Oxford, with telephone backup available at all times (24/7, 365 days a year). The randomisation program will use a minimisation algorithm to ensure balance between the groups with respect to collaborating hospital, sex, multiple births, gestational age at birth and existing diuretic therapy for the 24 hours prior to randomisation. Babies from multiple births will be randomised individually.

A Senior Trials Programmer at the NPEU CTU will write the randomisation program and hold the treatment allocation codes. If necessary, the code may be broken for a single baby at the request of the site Principal Investigator (PI) or clinician in charge of the baby. See Section 4.5 for the procedure for unmasking treatment allocation.

4.5. Unmasking

In the event of an emergency, a participant may be unmasked by the clinician at site by logging in to the randomisation website using a single-use access code provided in a sealed envelope. The reason for unmasking must be recorded. Clinicians carrying out emergency unmasking must be satisfied that it is a genuine emergency and that knowledge of the treatment allocation (either dexamethasone or placebo) is needed to guide the appropriate clinical management of the

participant. In some cases this may be achieved without unmasking by treating the participant as if they have received dexamethasone.

As it is best practice not to unmask participants until any follow-up is completed, all other requests for unmasking must be made in writing to the NPEU CTU, who along with Chief Investigator and Principal Investigator will consider the request.

4.6. Cytokines

Parents of babies recruited at specified sites will be asked to consent to having opportunistic microsamples of blood and endotracheal tube secretions taken at study entry, and within the 4-hour period prior to the IMP dose on days 4, 7, 10 and 14 after randomisation.

A maximum of 0.5 ml of blood (total blood volume 1.5 to a maximum of 2.5ml over the 14 days) will be obtained as an "extra sample" by trained staff at the time of a clinically indicated capillary blood sampling episode on these 5 occasions. Endotracheal tube secretions will be obtained at the time of clinically indicated endotracheal tube suction (in ventilated babies only). The endotracheal tube suction will be performed by trained staff.

All local policies regarding infection prevention and analgesia will be adhered to throughout sample collection. Trained staff will then undertake preliminary sample processing on the neonatal unit, following which the samples (EDTA plasma and endotracheal tube secretions) will be frozen and stored on site at -80°C prior to transportation to the laboratory at St James's University Hospital for storage and subsequent analysis. The University of Leeds -80°C freezer facility is linked to a 24h temperature monitoring system (Tutela) with back-up freezer space available in case of freezer failure.

4.6.1. Cytokine profiling

Cytokines will be profiled in plasma and endotracheal tube secretions by multiplex immunoassay (Bio-Plex Pro; Bio-Rad) on a Luminex-200 cytometer equipped with Bio-Plex Manager software (version 6.11). The following multiplex immunoassay kits will be used: Bio-Plex Pro Human Cytokine 21-plex Assay #MF0005KMII (analytes: IL-1α, IL-2Rα, IL-3, IL-12 (p40), IL-16, IL-18, CTACK, GRO-α, HGF, IFN-α2, LIF, MCP-3, M-CSF, MIF, MIG, β-NGF, SCF, SCGF-β, SDF-1α,TNF-β, TRAIL) and Bio-Plex Pro Human Cytokine 27-plex Assay #M500KCAF0Y (analytes: FGF basic, Eotaxin, G-CSF, GM-CSF, IFN-γ, IL-1β, IL-1ra, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12 (p70), IL-13, IL-15, IL-17, IP-10, MCP-1 (MCAF), MIP-1α, MIP-1β, PDGF-BB, RANTES, TNF-α, VEGF). Plasma samples will be diluted according to the manufacturer's recommendations. For the endotracheal tube secretions, a small aliquot (20µl) will be set aside for protein concentration determination (see below). The remainder will be diluted 50:50 in 0.5% bovine serum albumin in phosphate buffered saline prior to multiplex immunoassay profiling. All samples (50µl) will be assayed singly (due to limited available sample volume) according to the manufacturer's recommended protocol and plates washed using a Bio-Plex handheld magnetic washer (#171-020100; Bio-Rad). Plates will be run on the pre-calibrated Luminex-200 cytometer and data analysed using Bio-Plex Manager software. Cytokine profiles from plasma samples will be expressed as pg/ml whereas the endotracheal secretions will be standardised to total protein concentration (pg/mg protein) due to anticipated variability in PBS flush volume. Total protein concentration of endotracheal secretions will be determined in triplicate using a modified Lowry assay (Bio-Rad DC assay #5000112) using the microtitre plate method, following the manufacturer's instructions. All data will be stored on a secure server at the University of Leeds.

4.6.2. Assay QC considerations

The Luminex-200 machine is serviced annually and will be validated on the day prior to running the assay (Bio-Plex Validation Kit 4.0 #171203001, Bio-Rad). On the day of the assay, the machine is pre-calibrated using the Bio-Plex Calibration Kit #171203060, Bio-Rad). Validation outputs will be stored on a secure server at the University of Leeds. All assays will be run on the same day and using the same lot number of assay kit. Intra and inter-plate assay precision (% coefficient of variation) will be assessed using a validation sample (normal human adult EDTA plasma from a single donor), which will be assayed in triplicate on each plate. Kits will be stored at 4ºC in a cold room equipped with a temperature monitoring system (Tutela). Intra and inter-assay precision data will be stored along with assay date and kit lot number and expiry date on a secure server at the University of Leeds. Samples will be thawed only once and immediately prior to running the assay. All protein assays will be carried out on the same day as each other with the same standard dilution series, ensuring samples are diluted to fall within the linear range of the standard calibration curve (bovine serum albumin, provided with the kit). An internal standard (a stock of BSA of known concentration) will be run in triplicate on each assay plate to assess intra and inter-plate assay precision. Reconstituted calibration and internal standard stocks will be aliquoted for single use and stored in a temperature monitored -80°C freezer. A dedicated protein assay kit will be used for trial samples and the expiry date and lot number will be recorded on the secure server along with the sample readings and %CVs. All research staff at the University of Leeds laboratory have up-to-date GCLP training.

4.7. Stopping Trial Medications

Trial medications will be stopped under the following circumstances:

1. parental wish
2. intolerable adverse reaction to study drug as determined by the treating clinician (e.g. anaphylaxis)
3. requirement for cyclooxygenase therapy
4. if it is deemed by the treating clinician to be in the baby's best interest (e.g. overwhelming sepsis or need for open label corticosteroid).

If the trial medication is stopped, data should still be collected for the remainder of the trial. No further blood and bronchoalveolar samples should be taken if the baby is participating in the cytokine sub-study. If samples are taken inadvertently after the IMP is stopped, these can still be analysed in the study.

If parents choose to withdraw their baby from the trial, permission will be sought to continue data collection and/or use data up to the point of withdrawal from the trial.

The reason for withdrawing the baby will be recorded in the CRF and medical notes. As the babies are hospital inpatients, clinical monitoring will be ongoing.

4.8. Definition for the End of Trial

The date of the database lock.

5. Investigational Medicinal Product

5.1. Dosing and Administration

Dexamethasone (Hospira, dexamethasone 3.3 mg/ml solution for injection) will be supplied as a clear sterile solution at a concentration of 3.3 mg per 1 ml in 2 ml vials. Cartons containing 14 single-use vials will be provided. Each carton will be labelled with a unique code and in compliance with the guidance given in Annexe 13 of the European Commission's guidelines for Good Manufacturing Practice.

Doses of 50 mcg/kg (0.015 ml/kg of 3.3 mg/ml solution) of dexamethasone will be administered daily on days 1 to 10 after randomisation (10 doses) then alternate days on days 12, 14 and 16, making a total of 13 doses. Doses are to be calculated on the current working weight of the baby at the time of randomisation (or may be increased in line with current working weight as per local practice) and diluted to appropriate volume with dextrose or saline. Current working weight will be that which is designated by individual units as the weight to manage IMPs with. While the baby has an intravenous line in situ (peripheral or central) each dose is to be given as a short intravenous infusion over 10–15 minutes. Once the baby has had intravenous access removed the IMP may be given via the nasogastric tube or orally. The dose should be prepared and diluted in the same manner for oral, nasogastric and intravenous administration.

All 13 doses will be given unless there are adverse effects necessitating stoppage, as referenced in Section 4.7. Placebo will be supplied as a clear sterile solution of 0.9% saline solution for injection. Cartons identical to those for dexamethasone, each containing 14 identical single-use vials will be provided. Volume of IMP to be withdrawn from the vial will be calculated following the calculations for dexamethasone dosing and then diluted with dextrose or normal saline for administration.

Following randomisation, first dose should be administered after the necessary baseline data collection. Once the vial is opened the drug must be used immediately.

5.2. Summary of Product Characteristics (SmPC)

The Reference Safety Information for pharmacovigilance purposes will be the SmPC for the dexamethasone 3.3 mg/ml product manufactured by Hameln.

5.3. Storage of IMP

The IMP will be stored in accordance with the SmPC.

5.4. Distribution

Sufficient supplies of IMP will be provided to each site. Distribution and use of IMP will be tracked by the staff at the NPEU CTU using an online web-based pack management system. Additional supplies will be provided as and when needed.

5.5. Accountability

Trial drug packs will be received by pharmacy and stored on the neonatal units. Detailed accountability records will be maintained using an electronic stock management system to document which pack of medication is dispensed to each baby. Site staff will be required to write the baby's trial number and initials on the trial pack allocated.

Pharmacy will maintain an overall inventory of stock on a paper log.

5.6. Background Medication

Caffeine therapy is an inclusion criterion (please refer to SmPC of relevant product used at site).

5.7. Open-Label Treatment

If the clinical condition of a baby warrants intervention, treatment with open-label corticosteroids after the intervention period may be given. Clinical responsibility for the care of the baby will remain fully with the neonatal clinical team irrespective of the trial.

Use of open-label treatment will routinely be recorded.

5.8. Special Warnings and Precaution for Use with Concomitant Medications

Should a baby develop a requirement for cyclooxygenase therapy, develop overwhelming sepsis, or require treatment which would be contra-indicated with use of steroids in the opinion of the treating clinical team, the study medication should be stopped but follow-up assessments should continue.

6. Pharmacovigilance

6.1. Definitions

Adverse Event (AE)	Any untoward medical occurrence in a participant to whom a medicinal product has been administered, including occurrences which are not necessarily caused by or related to that product.
Adverse Reaction (AR)	An untoward and unintended response in a participant to an investigational medicinal product which is related to any dose administered to that participant. The phrase "response to an investigational medicinal product" means that a causal relationship between a trial medication and an AE is at least a reasonable possibility, i.e. the relationship cannot be ruled out. All cases judged by either the reporting medically qualified professional or the Sponsor as having a reasonable suspected causal relationship to the trial medication qualify as adverse reactions.
Serious Adverse Event (SAE)	A serious adverse event is any untoward medical occurrence that: • results in death • is life-threatening • requires inpatient hospitalisation or prolongation of existing hospitalisation • results in persistent or significant disability/incapacity • consists of a congenital anomaly or birth defect. Other 'important medical events' may also be considered serious if they jeopardise the participant or require an intervention to prevent one of the above consequences. NOTE: The term "life-threatening" in the definition of "serious" refers to

	an event in which the participant 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.
Serious Adverse Reaction (SAR)	An adverse event that is both serious and, in the opinion of the reporting Investigator, believed with reasonable probability to be due to one of the trial treatments, based on the information provided.
Suspected Unexpected Serious Adverse Reaction (SUSAR)	A serious adverse reaction, the nature and severity of which is not consistent with the information about the medicinal product in question set out: • in the case of a product with a marketing authorisation, in the summary of product characteristics (SmPC) for that product • in the case of any other investigational medicinal product, in the investigator's brochure (IB) relating to the trial in question.

NB: to avoid confusion or misunderstanding of the difference between the terms "serious" and "severe", the following note of clarification is provided: "Severe" is often used to describe intensity of a specific event, which may be of relatively minor medical significance. "Seriousness" is the regulatory definition supplied above.

6.2. Causality

The relationship of each adverse event to the trial medication must be determined by a medically qualified individual according to the following definitions:

Unrelated – where an event is not considered to be related to the IMP

Possibly – although a relationship to the IMP cannot be completely ruled out, the nature of the event, the underlying disease, concomitant medication or temporal relationship make other explanations possible.

Probably – the temporal relationship and absence of a more likely explanation suggest the event could be related to the IMP.

Definitely – the known effects of the IMP, its therapeutic class or based on challenge testing suggest that the IMP is the most likely cause.

All AEs (SAEs) labelled possibly, probably or definitely will be considered as related to the IMP.

The final decision relating to causality must be made by a medically qualified Investigator who is a member of the study team.

6.3. Recording SAEs

Non-serious adverse events will not be routinely recorded as the IMP is well-studied in this age group with fairly extensive reporting of safety outcomes in systematic reviews. Also, the IMP is a licenced product which is being given at a much lower dose than the standard dose for neonates. However, adverse events which are part of the safety outcomes of the trial will be recorded in the CRF. SAEs as described in this section will be monitored from randomisation until 14 days after last dose of treatment with the IMP and recorded on the CRF.

6.4. Reporting Foreseeable Serious Adverse Events

The following are serious adverse events that could be reasonably anticipated to occur in this population of infants during the course of the trial or form part of the outcome data. They do not require reporting by the trial centres as SAEs but do require relevant data to be captured in the CRF:

- Death (unless cause not anticipated in this population)
- Necrotising enterocolitis or gastrointestinal perforation
- Bronchopulmonary dysplasia (or chronic lung disease as these are the same)
- Intracranial abnormality (haemorrhage or focal white matter damage) on cranial ultrasound scan or other imaging
- Pulmonary haemorrhage
- Pneumothorax
- Anaemia requiring blood transfusion
- Hyperbilirubinaemia
- Hyperglycaemia
- Coagulopathy requiring treatment
- Hypotension
- Hypertension
- Impaired renal function
- Patent ductus arteriosus (PDA)
- Retinopathy of prematurity
- Sepsis
- Fractures
- Clinically significant liver failure
- Clinically significant left ventricular hypertrophy on echocardiography
- Hydrocephalus

Only if these events are thought to be causally related to the IMP would they require urgent reporting to the trial centre as outlined in Sections 6.5 and 6.7.

6.5. Reporting All Other SAEs

All other SAEs will be reported by trial sites to the NPEU CTU immediately, at least within 24 hours of the site staff becoming aware of the event. If an individual has OpenClinica access, an electronic SAE form should be signed and submitted. If not, a paper SAE form should be completed and faxed or emailed to the NPEU CTU. If a telephone call is made, site staff will follow up this notification with a SAE report form on OpenClinica (if an individual has access) or by fax/email as soon as possible but no later than 24 hours after becoming aware of the event.

Additional information received for a case (follow-up or corrections to the original case) needs to be detailed on a new SAE form and faxed or emailed to the NPEU CTU. Guidance outlining the reporting procedure for clinicians will be provided with the SAE form and in the trial handbook.

NPEU CTU will review the report, request any additional information and ensure it is assessed by the Chief Investigator (CI, or their delegate) within the reporting timeframe. It will also be reviewed at the next Data Monitoring Committee (DMC) meeting. The CI will inform all Principal Investigators of relevant information that could adversely affect the safety of the participants.

6.6. Expectedness

Expectedness will be determined according to the SmPC for Caffeine and Dexamethasone respectively.

6.7. Reporting SUSARs

Treatment codes will be unblinded for specific participants and SUSARs will be reported if the participant received the active product or a SUSAR is suspected to be related to a component of the placebo.

All SUSARs will be reported by NPEU CTU to the relevant Competent Authority (MHRA in the UK), the chair of the DMC, and to the REC and other parties as applicable. For fatal and life-threatening SUSARs, this will be done no later than 7 calendar days after the NPEU CTU is first aware of the reaction. Any additional relevant information will be reported within 8 calendar days of the initial report. Any other SUSARs will be reported within 15 calendar days.

6.8. Safety Monitoring

During the course of the trial, safety data will be reviewed by the DMC. This will include safety data for SAEs as stated in Sections 6.4-6.7. The DMC will, if appropriate, make recommendations regarding continuation of the trial or modification of the trial protocol. The Trial Steering Committee (TSC) will have ultimate responsibility for deciding whether the trial should be stopped on safety grounds.

6.9. Development Safety Update Reports

The CI will submit (in addition to the expedited reporting above) Development Safety Update Reports (DSURs) annually throughout the trial, or on request to the Competent Authority, Ethics Committee, Host NHS Trust and Sponsor.

7. Statistical Considerations

7.1. Sample Size / Power Calculations

For the primary outcome of time to extubation, assuming 90% power and two-sided 5% level of significance, a log rank test with a sample size of 70 babies (35 in each arm) would be required to detect a HR of 2.3 when the control group hazard rate is 1.0. This HR is the lower end of the confidence interval found by our retrospective study; adjusted HR 6.24 (95% CI 2.34 to 16.63)[17]. This sample size will allow the detection of a clinically important absolute risk reduction of 36% for the secondary outcome of failure to extubate by day 7, with 90% power, a two-sided 5% level of statistical significance, and a control group incidence of 80%, (consistent with control group event rates reported in previous trials[15,17]). As other previously published randomised trials of dexamethasone report failure to extubate by day 7 as a primary outcome, we consider this an important secondary outcome to consider in the sample size calculations.

Since all centres routinely collect the required data we anticipate a negligible loss to follow-up. However, as the primary analysis will be per-protocol, babies will be censored for the primary outcome (and other time-to-event outcomes) if the baby either dies, discontinues treatment before extubation, or remains intubated at day 16 (last day of the intervention period). These babies will be excluded from the analysis of other secondary outcomes.

Studies and published data sets show that 13.5–18% of babies <30 weeks' gestational age will have late onset sepsis,[2,18] 7% will be treated for NEC,[1] and 14% will be treated for PDA with cyclooxygenase inhibitors.[19] We estimate that a third of late onset sepsis, half of NEC and a quarter of treated PDA would occur in the study period. We therefore expect that up to 14% of babies will permanently discontinue the study treatment for clinical reasons. We estimate that 10% of babies will either die before extubation, or remain intubated at day 16. These deaths will be reported to the DMC who will monitor for any safety imbalances or differences in censoring rates between the groups. The per-protocol analysis will therefore censor or exclude approximately 23% of all babies giving a final overall sample size target of at least 90 babies.

7.1.1. Cytokine profile

With regard to the secondary outcome of inflammatory network modelling, the samples obtained from specified sites (approximately 16 babies) will be ample for detecting differences in absolute cytokine concentrations. This assumption is based on work conducted to date in the assisted conception setting[20,21] rather than power calculations since the principal purpose of measuring cytokines in the current context is not to determine changes in individual cytokines, but rather to understand differences in network evolution in relation to response to dexamethasone and those characterising BPD.

7.2. Hypotheses to be tested

To determine if the treatment of ventilator dependent preterm babies of less than 30 weeks' gestation who are at high risk of developing BPD with very low dose dexamethasone impacts upon:

i. Time to extubation (where the participant remains extubated for more than 24 hours)
ii. Time to extubation (whether or not the participant remains extubated for more than 24 hours)
iii. Rates of extubation by day 7 (where the participant remains extubated for more than 24 hours)
iv. Rates of extubation by day 7 (whether or not the participant remains extubated for more than 24 hours)
v. Survival to 36 weeks' PMA (or discharge home if sooner)
vi. Respiratory morbidity to 36 weeks' PMA (or discharge home if sooner)
vii. Cytokine profile
viii. Safety outcomes
ix. Parent/family experience

7.3. Statistical Analysis

Demographic factors, clinical characteristics, and service utilisation will be summarised with counts (percentages) for categorical variables, mean (standard deviation (SD)) for normal distributed variables or median (interquartile or other percentile range) for other continuous variables.

Comparative statistical analysis will entail calculating the HR plus 95% confidence interval (CI) for the primary outcome, the risk ratio (plus 95% CI) for dichotomous outcomes, and mean or median difference (plus 95% CI), for other continuous variables. Since this is an efficacy study, for the primary analysis, babies will be analysed on a per-protocol basis, excluding babies who are discontinued from the study treatment. For the primary outcome events will be censored for babies who discontinue or die before extubation or who remain intubated at day 16.

A secondary analysis will analyse all outcomes on an intention-to-treat basis. The consistency of the effect of dexamethasone will be explored to see whether it is of particular help to specific subgroups of babies, using the statistical test of interaction. The following pre-specified subgroups will be examined for the primary outcome (1) gestational age, (2) singleton vs. multiple pregnancy and (3) diuretic use at trial outset.

Basic cytokine data analysis will be largely conducted using a combination of Kruskal-Wallis and post hoc Dunn's tests. Significance levels will be corrected for multiple comparisons by applying Holm's sequentially selective Bonferroni method. Thereafter, correlation, principal components and factor analyses will be used as recognised tools to investigate mediator interrelationships. More informative network-based approaches will be used to model inflammatory cytokine:cytokine network interactions using Bayesian methods refined in-house. These networks offer a framework combining uncertainty (probabilities) and logical structure (independence constraints) to represent complex biological phenomena by generating acyclic graph-based models of joint probability distributions representing the conditional independence between related variables. The key element of this approach is that this allows investigators to capture the complex biological features of cytokines such as antagonism, synergy and functional redundancy, as we have recently shown (Field et al., BMC Systems Biology, In Press) rather than relying on the increasingly inadequate T helper cell type 1/2 (Th1/Th2) paradigm.

Two dichotomisations will be applied: one in response to dexamethasone therapy versus placebo and one relating to the time to extubation (groupings for the latter will be determined empirically based on the data emerging from the study), independent of treatment. Both will incorporate a dynamic aspect to the Bayesian model to account for changes in cytokine profiles and interrelationships over time. Data from each group will also be used to develop modified variational Bayesian state space models (where we will also use the human-specific text learning algorithm (https://compbio.dfci.harvard.edu/predictivenetworks/) to generate prior networks. Initialisation fields will be set to 10 seeds and 1,000 iterations, and the maximum hidden state dimension limited to 20. A z score equivalent to a 98% significance threshold will be used to define the number of edges reaching significance. Networks will be visualised using either Cytoscape or the open graph Viz platform Gephi. The spatialisation technique applied will be the force-directed Yifan Hu algorithm, whereas a community detection algorithm will be used to detect the underlying graph topology modular structure. We also plan to use machine learning-based methods in an attempt to identify the value of specific small groups of cytokines as predictors of outcome by using a support vector machines and random forest-based combinatorial approach developed in-house. The main focus of these will be to offer a starting point for an iterative stepwise empirical identification of outcome predictors based on feature importance ranking. Further modelling will be performed according to the manifestation of BPD as a secondary outcome measure.

8. Data Management

All trial data will be collected using bespoke CRFs. Data will be processed in line with the NPEU CTU Data Management standard operating procedures, using validated data management systems to ensure consistency, viability and quality of the data.

8.1. Source Data

Source documents are where data are first recorded, and from which participants' CRF data are obtained. These include, but are not limited to, hospital records (from which medical history and previous and concurrent medication may be summarised into the CRF), clinical and office charts, laboratory and pharmacy records, diaries, physiological data and correspondence.

CRF entries will be considered source data if the CRF is the site of the original recording (e.g. there is no other written or electronic record of data). All documents will be stored safely in confidential conditions.

8.2. Access to Data

Direct access will be granted to the research team, authorised representatives from the Sponsor, host institution and the regulatory authorities to permit trial-related monitoring, audits and inspections.

8.3. Data Handling and Record Keeping

The CI will take overall responsibility for ensuring that each participant's information remains confidential. All records will be stored securely and kept in strict confidence in compliance with the Data Protection Act. Minimal personal data will be collected and stored at NPEU CTU. Data collected will be stored in an electronic database in which the participant will be identified by a trial specific number. The infant's name and any other identifying details will be stored separately and linked by the trial number. This information will be stored for a period of no less than 25 years in order to follow up health related issues that may become relevant in the future. After the trial has been completed and the reports published, the data will be archived in a secure physical or electronic location with controlled access.

Storage will be on a restricted area of a file server. The server is in a secure location and access is restricted to a few named individuals. Offices where data are stored are located in a secure area via an electronic tag and individual rooms are kept locked when unoccupied.

Data will be processed on a workstation by authorised staff. The computer workstations access the network via a login name and password (changed regularly). No data are stored on personal workstations. Backing up is done automatically overnight to an offsite storage area. The location of the backup computer is in a separate department.

8.4. Archiving

In line with the principles of good clinical practice (GCP)/UK Clinical Trial Regulations guidelines, at the end of the trial, data will be securely archived at each participating centre in line with local policy. All essential documents will be kept for no less than 25 years in order to follow up health related issues that may become relevant in the future.

9. Serious Breaches

The Medicines for Human Use (Clinical Trials) Regulations contain a requirement for the notification of Serious Breaches to the MHRA within 7 days of the NPEU CTU becoming aware of the breach.

A serious breach is defined as a breach of GCP or the trial protocol which is likely to affect to a significant degree:

(a) The safety or physical or mental integrity of the subjects of the trial; or

(b) The scientific value of the trial.

In the event that a serious breach is suspected the NPEU CTU must be contacted as soon as practicable. In collaboration with the CI and the Sponsor, the serious breach will be reviewed by the NPEU CTU and, if appropriate, the NPEU CTU will report it to the REC committee and MHRA within 7 calendar days of the NPEU CTU becoming aware of the breach.

10. Trial Governance

10.1. Quality Assurance

The trial will be conducted in accordance with the current approved protocol, Good Clinical Practice (GCP), relevant regulations and standard operating procedures. The CI and PIs, with the support of the Project Management Group (PMG) will be responsible for the day-to-day smooth running of the trial at the site. They will encourage recruitment, provide staff education and training, and monitor data completeness and quality. The PMG will perform a trial risk assessment prior to commencement that will be reviewed at regular intervals during the course of the trial. The degree of central and site monitoring will be outlined in a separate Monitoring Plan, developed in light of any risks identified in the risk assessment. Site monitoring will be carried out by a suitably qualified person independent of the study team.

The NPEU CTU has systems in place to ensure that there is reporting and appropriate action taken in respect of:

(a) Serious breaches of GCP, the trial protocol and the Clinical Trial Authorisation

(b) Urgent safety measures

(c) Protocol violations.

Investigators will promptly notify the NPEU CTU, who will inform the Sponsor QA Office, of the following within the required timeframe, once they become aware of:

(a) Serious breaches of GCP, the trial protocol and the Clinical Trial Authorisation

(b) Urgent safety measures

(c) Protocol violations

(d) Any amendments to the trial

(f) Any other issues as stated in the Research Sponsorship Agreement (RSA).

The Sponsor reserves the right to audit any site involved in the trial and authorisation for this is given via the RSA.

10.2. Project Management Group

The trial will be supervised on a day-to-day basis by the PMG. This group reports to the Trial Steering Committee (TSC). At each participating centre, a local Principal Investigator will report to the PMG via the staff based at the NPEU CTU.

The core PMG will consist of the CI, Lead Clinician and NPEU CTU staff including:

- CTU Director
- Senior Trials Manager
- Senior Trials Programmer
- Trial Co-ordinator
- Trial Statistician
- Trial Programmer
- Administrator/Data Manager

The core PMG will meet regularly every 2 months throughout the trial. The core PMG will report to the Trial Steering Committee (TSC).

10.3. Co-Investigators group

The CIG will meet regularly to review progress, troubleshoot and plan future strategies. The CIG will comprise all co-applicants and the members of the core PMG.

10.4. Trial Steering Committee

The trial will be overseen by a TSC consisting of an independent chair and at least two other independent members. Committee members will be deemed to be independent if they are not involved in trial recruitment and are not employed by any organisation directly involved in the trial conduct.

Representatives from relevant Patient/Public Involvement groups, the Chief Investigator, other Investigators/co-applicants will be joined by observers from the NPEU CTU. The EME programme manager will be invited to attend all TSC meetings.

The role of the TSC is to provide the overall supervision of the trial. The TSC should monitor the progress of the trial and conduct and advise on its scientific credibility. The TSC will consider and act, as appropriate, upon the recommendations of the DMC and ultimately carries the responsibility for deciding whether a trial needs to be stopped on grounds of safety or efficacy.

10.5. Data Monitoring Committee

The DMC will be independent of the applicants and the TSC. It will review the progress of the trial and interim analysis at least annually, and provide advice and recommendations on the conduct of the trial to the TSC and (via the TSC) to the funders and the sponsor.

11. Ethical and Regulatory Considerations

11.1. Declaration of Helsinki

The trial will be performed in accordance with the recommendations guiding ethical research involving human subjects adopted by the 18th World Medical Assembly, Helsinki, Finland, 1964, amended at the 48th General Assembly, Somerset West Republic of South Africa, October 1996. Informed written consent will be obtained from the parents prior to randomisation/registration into the study. The right of a parent to decline their baby's participation without giving reasons must be respected. The parents will remain free to withdraw at any time from the study without giving reasons and without prejudicing their baby's further treatment. The study will be submitted to and approved by a main Research Ethics Committee (REC) and the appropriate Research Ethics Committee (REC) for each participating centre prior to entering babies into the study.

11.2. Good Clinical Practice (GCP) and Regulatory Compliance

This clinical trial, which involves the use of an investigational medicinal product has been designed and will be run in accordance with the principles of GCP and the current regulatory requirements, as detailed in the Medicines for Human Use (Clinical Trials) Regulations 2004 (UK S.I. 2004/1031) and any subsequent amendments of the clinical trial regulations.

11.3. Approvals

The protocol, informed consent form, Parent Information Leaflet and any proposed advertising material will be submitted to an appropriate Research Ethics Committee (REC), regulatory authorities (MHRA in the UK), and host institution(s) for written approval.

The Investigator will submit and, where necessary, obtain approval from the above parties for all substantial amendments to the original approved documents.

11.4. Reporting

11.4.1. Annual Reports

The CI shall submit once a year throughout the clinical trial, or on request, an Annual Progress Report describing the general progress and any relevant safety data to the REC, host organisation, and Sponsor.

11.4.2. End of Trial Report

At the end of trial (defined in 4.8), an end of trial notification and final report will be submitted to the MHRA, the REC, host organisation and Sponsor.

11.5. Participant Confidentiality

The trial staff will ensure that the participants' confidentiality is maintained. Minimal personal data will be collected and stored at NPEU CTU. Data collected will be stored in an electronic database in which the participant will be identified by a trial specific number. The infant's name and any other identifying details will be stored separately and linked by the trial number. All documents will be stored securely and will only be accessible by trial staff and authorised personnel. The trial will comply with the Data Protection Act.

12. Finance

12.1. Statement of Indemnity

Clinical negligence indemnification will rest with the participating NHS Trust or Trusts under standard NHS arrangements.

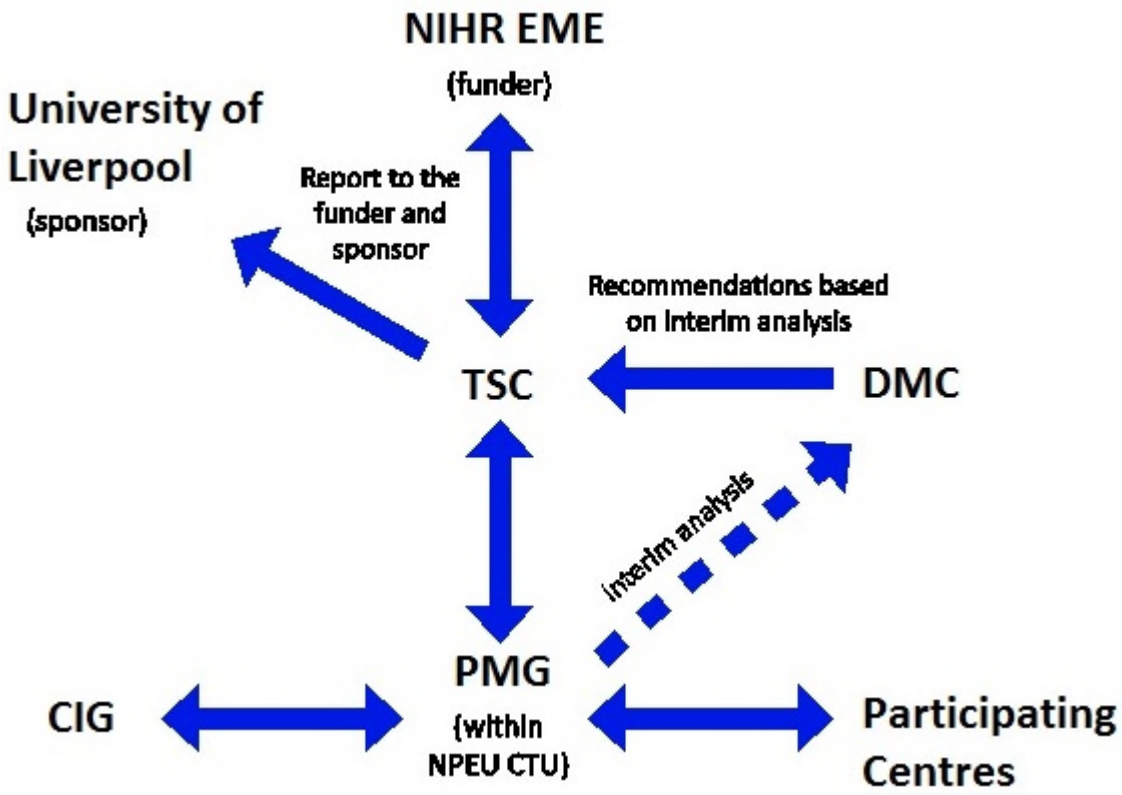
12.2. Funding

The National Institute for Health Research (NIHR) Efficacy and Mechanism Evaluation (EME) programme is funding the trial. The funding for the cytokine analysis is provided by the Children's Charity Cerebra.

13. Publication Policy

The success of the trial depends on a large number of neonatal nurses, neonatologists, and parents. Credit for the trial findings will be given to all who have collaborated and participated in the trial including all local co-ordinators and collaborators, members of the trial committees, the NPEU CTU and trial staff. Authorship at the head of the primary results paper will take the form "[name], [name] and [name] on behalf of the 'The Minidex Collaborative Group'". The drafting of the paper will be the responsibility of a writing committee. All contributors to the trial will be listed at the end of the main paper, with their contribution identified.

14.Study Organisational Structure



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16. Appendices

16.1. Appendix 1: Severity Based Diagnostic Criteria for BPD

Time point of assessment: 36 weeks' PMA	
Therapy with oxygen > 21% ***and/or*** respiratory support for ≥ 28 days cumulatively ***and*** the following levels of oxygen requirement as determined by the oxygen reduction test	
Mild BPD	Baby is breathing room air
Moderate BPD	Baby is in 22–29% oxygen, or 0.01–1.0 l/min
Severe BPD	Baby is in FiO_2 ≥ 0.3 or low flow oxygen ≥ 1.1 l/min or the baby is receiving respiratory support (invasive or non-invasive) to achieve saturations of ≥ 91%

The need for oxygen will be determined by the oxygen reduction test. This is based on the minimum inspired oxygen threshold at which the baby can maintain saturations of ≥ 91%. Babies requiring inspired oxygen to maintain saturations of ≥ 91% are considered to be oxygen dependent.

The test only applies to babies in < 0.3 inspired oxygen, or low flow oxygen of < 1.1 l/min, and who have not had respiratory support in the previous 24 hours. Those babies not meeting these criteria will not be tested, but their oxygen requirements captured on the relevant data form.

16.2. Appendix 2: Oxygen Reduction Test