CLINICAL INVESTIGATION PLAN

Official title: Paracetamol And Ibuprofen/Indomethacin in closing patent ductus arteriosus of preterm infants – randomised, placebo-controlled, multicentre trial (PAI)

Unique Protocol ID 38/2018
EudraCT ID 2018–000565-36
ClinicalTrials.gov ID NCT03648437

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Content

Signatures

The Principal Investigators have discussed this Clinical Investigation Plan (CIP). The Investigator agrees to perform the investigations and to abide by this CIP. The investigator agrees to conduct the investigation in compliance with the approved CIP, EU Good Clinical Practice (GCP), and other regulatory requirements.

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Signature Date

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1 Summary

Present randomised, placebo-controlled, double blind, phase 3, international, multicentre clinical trial aims to study the efficacy, safety, and pharmacokinetics of the combination of intravenous paracetamol vs. placebo (0.45% saline solution) with the standard prostaglandin synthase inhibitor medication, ibuprofen or indomethacin, in closing of patent ductus arteriosus (PDA) of preterm infants. The primary outcome is the ductal closure after the study drug. Preterm infants born before 37+0 gestation weeks and diagnosed to have a hemodynamically significant PDA, and who, according to the decision of the attending clinician, need the ibuprofen or indomethacin therapy, are eligible to this trial. The sample size to detect 41% increase in the number of infants with successful PDA closure (20% power, 5% alpha), n=30/group is needed. All patients are screened for their ductal status by cardiac ultrasounds, and possible adverse reactions. Additionally, patients' cerebral oxygenation (near infra-red spectroscopy), diuresis, and paracetamol serum levels are monitored. Infants will be recruited in four trial sites, three in Finland (Oulu, Helsinki, Turku), and one in Estonia (Tartu).

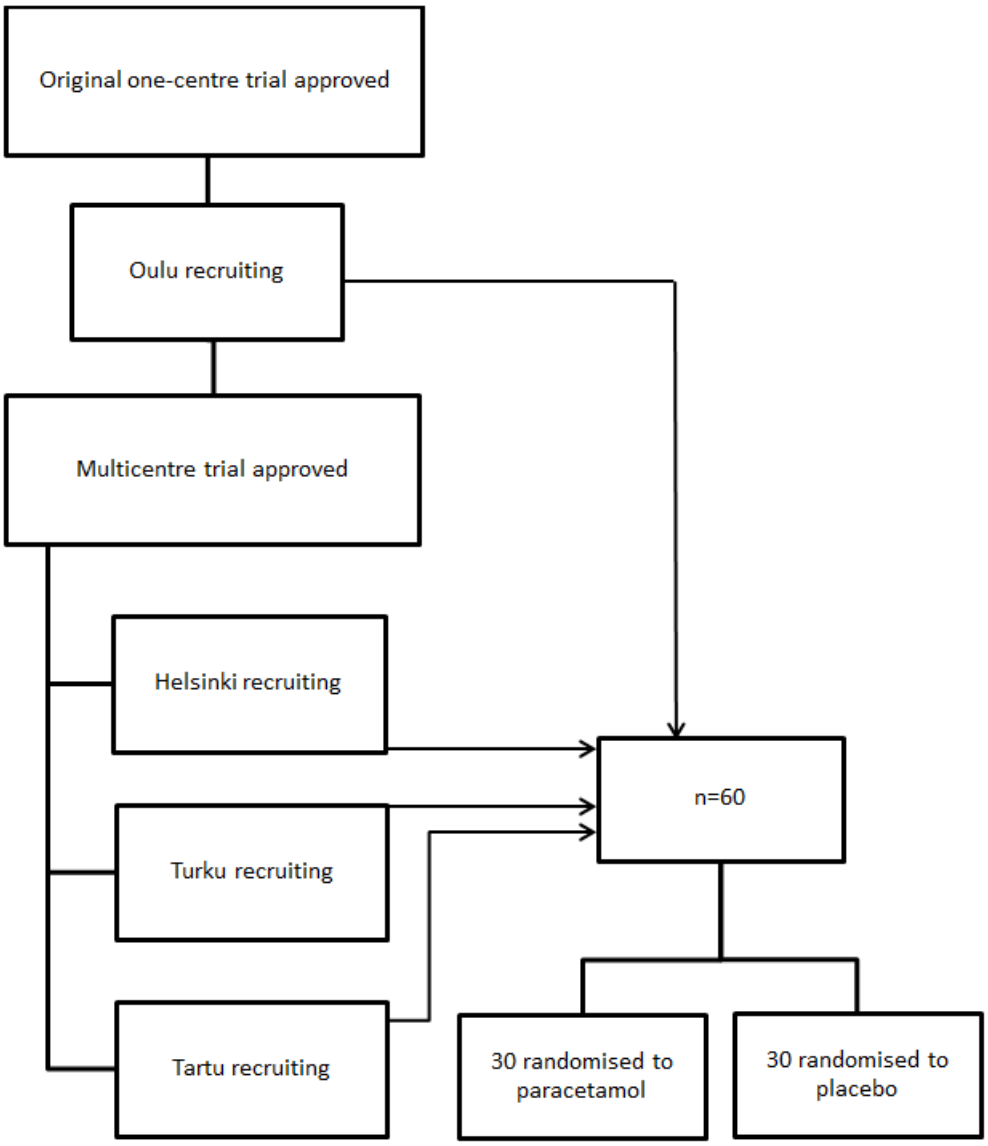

Figure1. Flow chart of the trial.

2 Background

Patent *ductus arteriosus* (PDA) may harm the recovery of very low gestational age preterm infants (VLGA, gestational age <32 weeks, or birth weight <1500 g).[1] Abundant blood flow from aorta *via* ductus to pulmonary and mesentery circulation increases blood pressure labiality and pulmonary oedema, decreasing cerebral oxygenation.[2] Constantly open ductus of a preterm infant has been associated to increased risks of bronchopulmonary dysplasia (BPD), necrotising enterocolitis (NEC), and intracerebral haemorrhage (ICH).[1, 3] Preterm infants with PDA had an eight-fold mortality compared to those with closed ductus.[4]

Although 34 % of small preterm infants have their ductus spontaneously closed, 55 % of those with birth weight less than 1000 g, and 60-70 % of those born before 28 gestation weeks, are estimated to receive therapy for persistent ductus arteriosus.[1]

2.1 Current PDA therapies

Non-steroidal anti-inflammatory drugs (NSAIDs), prostaglandin H_2 synthetase (PGHS) inhibitors, indomethacin and ibuprofen have been established as the standard medications for PDA.[5, 6] Surgical ligation of PDA is one possible choice for therapy.[1, 7] Both of these medications, as well as operative management, have harmful side effects in premature infants.[8]

Intravenous (IV) paracetamol has been used as a therapeutic drug for premature infants' pain medication.[9] In a previous observation study, five premature infants with hemodynamically significant PDA had their ductus closed by oral paracetamol.[10] The biological effect of paracetamol on the closure of ductus arteriosus was shown in a controlled, double-blind, phase 2 trial.[11] Lately, paracetamol has been shown to be as effective as PGHS inhibitors but better tolerated in closing PDA.[12]

Some very premature infants have remained devoid of the efficacy for ductal closure with ibuprofen or indomethacin.[13] Furthermore, even the effect of paracetamol in PDA closure was not detectable in the most premature infants born before 28 gestation weeks.[11] Due to the side effects, increasing dosage of PGHS inhibitors has not been considered acceptable.

2.2 Paracetamol mechanism of action

Paracetamol, ibuprofen and indomethacin inhibit the prostaglandin synthesis, but their mechanisms of action are not exactly the same.[14, 15] PGHS enzyme forms prostaglandins from the arachidonic acid. This enzyme consists of two compartments, oxygenase (COX) and peroxidase (POX). Ibuprofen and indomethacin inhibit the action of the COX compartment by competing of the binding site with arachidonate. Paracetamol impacts on the peroxidase compartment of the enzyme: it interferes with the chain reactions that produce essential co-substrate (critical tyrosine residue oxidation to a tyrosine phenoxyl radical) for COX activity and prostaglandin synthesis. (Figure 2).[16-19]

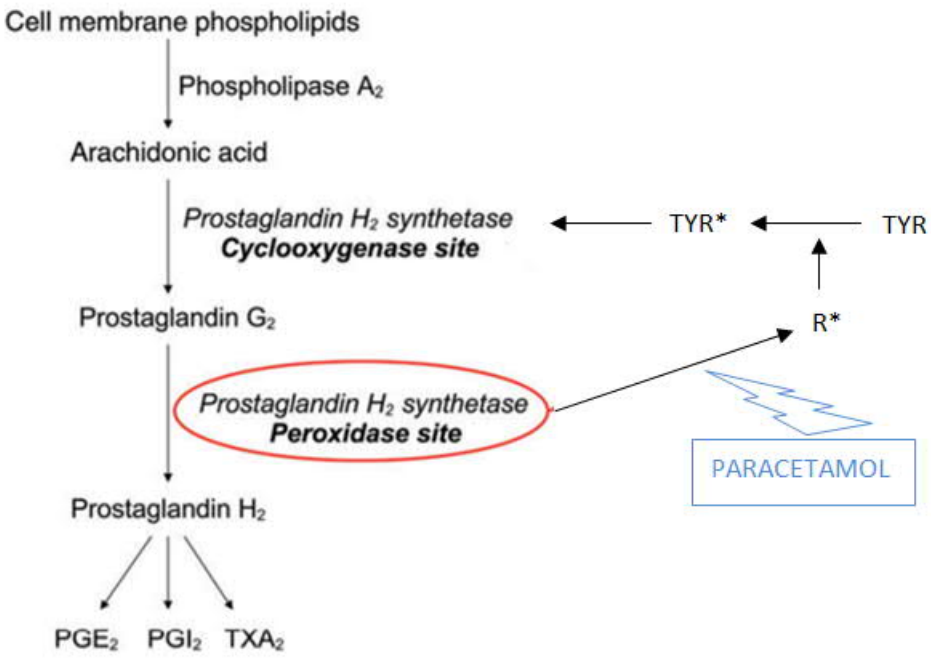

Figure 2. The suggested mechanism of action of paracetamol on the prostaglandin synthesis (TYR = tyrosine385; R = ferryl protoporphyrin IX radical cation).[18, 19]

2.3 Preliminary studies of combined PDA therapy with prostaglandin synthase inhibitors and paracetamol

For treatment of pain and fever, paracetamol and ibuprofen have been combined and studied in adults and children.[20-23] These combination preparations have been found well tolerated with strong safety profiles.

During ongoing PDA therapy by ibuprofen medication, the combined effect of coincidental, simultaneous paracetamol pain medication was studied in a cohort of premature infants.[24] VLGA infants born from January 2002 to December 2015 in Oulu University Hospital were studied. Altogether 113 VLGA infants were treated with ibuprofen. Of them, 18 had received concomitant IV paracetamol treatment for pain relief by chance. The first paracetamol dose was given at mean (SD) 1.6 (2.3) days age. Before the PDA treatment by ibuprofen, the mean (SD) cumulative paracetamol dose was 114 (83) mg/kg, and the paracetamol cumulative dose during ibuprofen was 71 (169) mg/kg. These infants were compared with 95 VLGA infants who had ibuprofen for PDA but no paracetamol. Infants with combination therapy tended to require less frequently the repeat ibuprofen dosages and/or ligations (27.8 vs 45.3%; p=0.056). No adverse reactions were detected. Neonatal outcomes were similar. IV paracetamol, given simultaneously with ibuprofen, was associated with tendency to fewer additional PDA therapies, potentially enhancing the ductal closure.

After this small cohort, two small pilot studies about combined ibuprofen and paracetamol therapy for PDA treatment in preterm infants have been published.[25, 26] The first one reported a randomised trial with 24 VLGA infants randomised to IV ibuprofen with paracetamol or IV ibuprofen with placebo.[26] They found a trend towards higher PDA closure rate in the paracetamol group (83% vs.

42%, p=0.08). The second one studied a series of 12 infants with monotherapy-resistant PDA.[25] Nine of them (75%) responded to combination therapy, avoiding ligation.

2.4 Safety of paracetamol

Traditionally, paracetamol has been marketed as a well-tolerated drug, and it is recommended even during pregnancy.[15] In a series of 189 neonates, including 62 preterm infants, no elevated liver enzymes prior to drug administration, during, or after administration was observed.[27] In a series of nine preterm infants (H25-31), the hepatic transaminase enzyme (AST) remained low for several days after medication (median 20 U/l, range 12 -186 U/l).[28] In addition, in a series of 50 newborn infants, intravenous paracetamol did not cause liver toxicity.[29] Paracetamol has not been found to cause any bleeding disorders, gastro-intestinal problems, or kidney failure. However, it can cause liver damage if repeated in very large doses.[30] On the other hand, according to some patient reports, no liver damage was evident even if mistakenly given overdoses of paracetamol up to 136 mg/kg, 146 mg/kg, or 446 mg/kg.[31-33]

In present project, our aim is to provide data towards a new management practice, i.e. advancing more effective closure of PDA before it causes adverse hemodynamic effects or requires treatments that are associated with serious adverse effects. This may be possible in case the treatment increases the closure of PDA, and the treatment is not associated with any evidence of serious adverse effects.

3 Trial objectives

3.1 Aim, primary and secondary outcomes

The purpose of this trial is to study the efficacy and safety of simultaneous intravenous ibuprofen/indomethacin and paracetamol medications in the closure of PDA in preterm infants.

Primary outcome

The primary outcome of this study is ductal closure after the study drug. The cardiac ultrasound examinations for the ductal status will be carried out at least before and after the study drug, and preferably at the discharge from the neonatal intensive care unit (NICU) whenever possible.

Secondary outcomes

Secondary outcomes include ductal closure time, the need for additional PDA therapies, cardiac ultrasound findings, including ductal calibres and LA/Ao ratios, laboratory values such as hepatic transaminases, renal function (cystatin-C), and paracetamol serum levels, daily diuresis, measured near infra-red spectroscopy (NIRS) values, and possible paracetamol adverse reactions.
Other secondary outcomes include the clinical patient data, the duration of any ventilation assist including the need for supplemental oxygen, the long term complications of prematurity (BPD

grades 2-3, ICH grades 2-4, NEC, retinopathy of prematurity needing therapy), other long-term morbidity, and mortality.

3.2 Hypotheses and objectives

Hypothesis:

1. Simultaneous IV paracetamol therapy with IV ibuprofen/indomethacin closes PDA in preterm infants.
2. Simultaneous IV paracetamol therapy with IV ibuprofen/indomethacin has no adverse effects in preterm infants.
3. The present paracetamol doses do not result in high paracetamol serum levels.

Objectives: to study in preterm infants requiring IV ibuprofen/indomethacin for treatment of PDA

1. efficacy of IV paracetamol with IV ibuprofen/indomethacin in a randomised, controlled, double blind clinical trial
2. safety of IV paracetamol IV ibuprofen/indomethacin in a randomised, controlled, double blind clinical trial
3. pharmacokinetics and pharmacodynamics of IV paracetamol

4 Methods

4.1 Study design

Present study is a randomised, placebo-controlled, double-blind, phase 3, international, multicentre clinical trial.

4.2 Sample size

The power calculation was based on the recent pilot trial where hemodynamically significant PDA closure rate was higher in the ibuprofen + paracetamol group than in the ibuprofen + placebo group, 83% vs. 42%, p=0.08.[26] In order to detect a 41% increase in the number of infants with successful PDA closure (80% power, 5% alpha), n=30/group is needed with the continuity correction included.

4.3 Study patients identification and criteria

Potentially eligible study patients will be identified during their NICU stay by members of the neonatal intensive care unit staff of the attending hospital sites. When a subject is identified and considered eligible for entry into this clinical investigation, the subject will be allocated the next available investigation number (subject ID number). This number will be the unique identifier of the

subject and noted on the CRF and all other documentation relating to that subject. Each subject that is enrolled into the study will have their study participation recorded in their hospital notes.

4.1.1 The inclusion criteria

Preterm infants born before 37+0 gestation weeks who are diagnosed to have a hemodynamically significant PDA, and who, according to the decision of the attending clinician, need ibuprofen or indomethacin therapy for it, are eligible to this trial.

Hemodynamically significant PDA definition:
Cardiac ultrasound findings may include large open ductus with calibre > 1.5-2 mm, left atrium to aorta (La/Ao) ratio >1,4, large shunt volume, ductal systolic flow rate <2 m/s, pulmonary artery's diastolic flow rate <30 cm/s, enlarged left ventricle, diastolic reflux in descending aorta.
Clinical picture includes symptoms such as worsening pulmonary function, otherwise unexplained tachycardia, diastolic hypotension; findings as typical murmur, hyperkinetic heart, active precordium, distending pulses; thorax x-ray findings as cardiac enlargement and increased oedema; other organ ultrasound findings as missing end-diastolic renal/cerebral/intestinal blood flow.

The patient attending to this trial is allowed to have a ventricular septal defect smaller than 3 mm in calibre, but bigger ones are regarded as the exclusion criteria. An open *foramen ovale* is considered physiological in preterm infants.

4.1.2 The exclusion criteria

Preterm infants with severe congenital malformations, suspected chromosomal defects, or suffering from any very severe life-threatening diseases (*e.g.* very severe birth asphyxia or persistent pulmonary hypertension, etc.) are not eligible to this trial.

4.1.3 Study open arm criteria

If the parents deny the consent for randomisation, they are asked for the permission for solely the use of the patient data of the child for research purposes. If this permission is given, these patients form the additional open arm of the study.

4.4 Randomisation and masking

The computed randomisation will be done beforehand by a separately nominated randomisation group. The trial code numbers are randomised and listed prior to the start of the trial.

The placebo drug is 0.45 % saline solution, similar to paracetamol, both being clear liquids. Therefore, the personnel will be blinded and remain unaware of which drug the patient receives. The study drug will be kept and prepared away from the NICU, at the separate office, in a locked cabinet. The study nurse, the pharmacist of the ward, or a nurse who does not participate in the study patients' treatment in any way, will prepare the study drug. They all are bound to secrecy about the study drug contents.

4.5 Informed consent process

The study patients will be recruited when the need for ibuprofen/indomethacin therapy is identified as soon as possible. The parents will be handed an information leaflet and a letter of invitation by the investigator at the site who will discuss with patients' parents for informed written consent (annex 1). Parents will have relevant time to consider the information. As the informed consent is given, the new study patient will be assigned a trial code number (ID) from the list made beforehand. The trial code number will match with a closed envelope where has been drawn a leaflet with the patient's study medication group written on it. The study medication will begin immediately.
If the parents deny the consent, the patient would be treated according to the standard PDA treatments of the attending hospitals (see 4.6).

4.6 Intervention

The study drug is continued for altogether three to six days, i.e. as long as the local standard PDA treatment period will last. The ductal outcomes are defined by the attending clinicians.

Standard PDA treatments include following ibuprofen or indomethacin medication courses or both, depending on the individual clinical scenario present:
Oulu, Helsinki, and Tartu: three days' ibuprofen therapy Pedea® 5mg/ml injection solution; dosing: 10 mg/kg + 5 mg/kg + 5 mg/kg q24h.
Turku: three days' indomethacin therapy 0.2mg/kg + 0,1mg/kg + 0,1mg/kg q24h.
Both of these courses may be repeated if the first one would not close PDA, the indomethacin course with doubled two last doses (0.2mg/kg). In case of any contraindications for ibuprofen or indomethacin, the treatment would be surgical ligation.

The masked study drug is either paracetamol 10mg/ml infusion solution (Paracetamol Fresenius Kabi 10mg/ml 50ml®), or placebo, half-physiological 0.45% saline (Natriumklorid 4.5mg/ml 500ml Braun®). The placebo is chosen in order to avoid the risk of hypernatremia. The study drug loading dose 20mg/kg will be given ± 6 hours from the first ibuprofen/indomethacin dose, and continued 7.5mg/kg every 6 hours up to 3 (-6) days. If the patient had received any paracetamol preparation within two days (48 h) before the study drug, no loading dose is given. The study drug is administered as 15 minutes infusion. The exact dates and time points of the administered study drug doses will be recorded.

No other paracetamol preparations are allowed to be given during the study drug. After the trial, the paracetamol administration should be limited if possible. Otherwise, two days wash-out period is recommended. All the other medical or other therapies will be prescribed by the judgment of the attending physicians. In cases of suspicion of severe side effects or other absolute necessities, the attending physician is allowed to break the study code, and stop the study drug (protocol violation).

The pain therapy needed is estimated using local neonatal pain scales and accomplished using openly the opioid or other preparations used normally in the each site (*e.g*. morphine, fentanyl).

The drug treatment continues for three days (12 doses), but may be repeated if the standard treatment course is repeated – loading dose will be left out if the patient had received paracetamol within 48 h before the 1st dose of the second course. The study drug treatment is to be discontinued, if the diagnosis of PDA requiring specific treatment is made during the drug treatment. Then discontinuing the study drug at least 8 hours before a specific PDA treatment should be considered.

4.6 NICU follow-up

The cardiac ultrasound examinations will be performed at least before the randomisation, and after the study drug course, and preferably at the discharge from the NICU whenever possible. The ductal status will be recorded and ductal calibres and if possible, LA/Ao-ratios will be measured.

During the study drug, diuresis (ml/kg/h) will be measured by weighing all the diapers during the study drug. These weights, the time points when measured, and the study patients' current weights will be recorded.

Whenever available, cerebral oxygenation by near infra-red spectroscopy (NIRS) will be measured at the forehead and values recorded.

The left-overs of the routine blood samples will be saved for paracetamol serum level measurements during the study drug. If available, the hepatic transaminases (P-/S-alat) and renal function (P-/S-cystatin C) will be measured the day after the study drug.

5 Safety

Previously, no hepatic or renal failure has been reported using the present study drug dosage. In our previous cohort studies and in the randomised trial of preterm infants, no signs of hepatic or renal insufficiency have been detected in any patient. [9, 11, 24, 34, 35]

The intravenously administered investigational product can cause no harm to study patients' families, nursing staff, or the ward milieu.

5.1 Adverse reactions: definitions, treatment and follow-up

All study infants will be observed for any clinical signs of adverse reactions of paracetamol. All adverse reactions are recorded in the medical records and CRF. The most severe adverse reaction would be severe hepatic or renal insufficiency that could be possible after an overdose. Any suspicion of hepatic or renal failure would be a reason to immediately stop the study drug and examine the serum transaminase levels. If necessary, the trial coding may be broken by one of the non-blinded staff members of the site. Symptoms of hepatic or renal failure are considered as an emergency.

Sudden unexpected severe adverse reactions (SUSAR) are recorded into a special file maintained by the Principal investigators. Of them, death or danger of life are reported to central contact person, who will report them to Fimea within seven days after the incident, others within 15 days. This file is reported to Fimea annually.

Possible adverse reaction symptoms may include

- Oliguria: decreased diuresis (<1ml/kg/h) due to renal insufficiency, as followed by weighing the diapers. The fluid restriction should be considered during the study drug course.
- Hypotension: blood pressure of preterm infants may decrease during loading dose of IV paracetamol. Blood pressure should be routinely measured during the study drug course.
- Skin colour: yellow. In order to detect possible hyperbilirubinemia, the serum or skin bilirubin levels should be measured during the study medication period. In case of exceptionally severe hyperbilirubinemia, an abdominal ultrasound scan should be done.

5.2 Paracetamol overdose: intoxication symptoms and treatment

Drug-induced liver damage (hepatotoxicity) does not result from paracetamol itself, but from one of its metabolites, N-acetyl-p-benzoquinoneimine (NAPQI). Therefore, in the case of overdose or intoxication, the possible symptoms would not manifest immediately after administration, but after a period of time. The hepatic injury can be prevented by using the available antidote, acetyl cysteine. Even the suspicion of paracetamol overdose should be taken seriously, the situation clarified and acetyl cysteine therapy started promptly.

Typical symptoms and signs of the paracetamol intoxication would be:

- During the first 24 hours after the drug administration, nausea, vomiting, weariness, lack of appetite, and sweating may occur.
- 24 – 48 hours after the administration, serum bilirubin and liver aminotransferase levels and prothrombin time increase. Diuresis may decrease due to dehydration (vomiting) or renal injury.
- 2 to 5 days after the administration the hepatic and renal failure is manifested, usually with metabolic acidosis.

Using early acetyl cysteine therapy, started earlier than 16 h after administration, the hepatic failure can be prevented, but if started later, the possible injury may be diminished as well. The intoxication treatment includes effective intravenous fluid therapy in order to attenuate the metabolic acidosis, and the diuresis should be aimed to be more than 1.5 ml/kg/h.

5.3 Adverse events: definitions and reporting

Adverse event (AE): Any untoward medical occurrence, unintended disease or injury, or untoward clinical signs (including abnormal laboratory findings) in subjects, users or other persons, whether or not related to the IP.

Serious adverse event (SAE): Any adverse event that led to death, serious deterioration in the health of the subject, that either resulted in a life-threatening illness or injury, or a permanent impairment of a body structure or a body function, or in-patient or prolonged hospitalisation, or medical or surgical intervention to prevent life-threatening illness, or injury or permanent impairment to a body structure or a body function.

An adverse event does not include:

- Medical or surgical procedures; the condition that leads to the procedure, is an adverse event.
- Pre-existing disease, conditions, or laboratory abnormalities present at the start of the study that do not worsen in frequency or intensity.
- Situations where an untoward medical occurrence has not occurred (e.g., hospitalizations for cosmetic or elective surgery or social/convenience admissions).
- The disease being studied or signs/symptoms associated with the disease unless more severe than expected for the subject's condition.
- Expected post-operative course.

All adverse events and SAEs will be recorded in the medical records and CRF.

Adverse events will be evaluated during 2-week period starting from the 1st study drug dose, using the PAI study adverse events -file. Each adverse event is classified by their possible association to the study drug. The classification scale is:
0 = no connection
1 = unlikely
3 = possible
4 = likely
5 = certain

All serious adverse events are reported to principal investigators and central contact person immediately, but no more than 3 calendar days after becoming aware of the event.
The Principal Investigator will complete the serious adverse event form and the form will be emailed to the central contact person to add, within 3 working day of his/her becoming aware of the event. The Principal Investigator will respond to any SAE queries raised by the central contact person as soon as possible. The central contact person will report to Fimea all reportable events within the specified timeframes.

In case of suspicion of severe safety issues associated with the trial, separately nominated safety group will be able to break the study code and interrupt the trial, if necessary.

5.4 Deviations from clinical investigation plan and withdrawal from the trial

A deviation is considered a departure from the conditions and principles of GCP in connection with that investigation; or the CIP relating to that investigation, as amended from time to time. The investigator shall not deviate from this CIP except in situations that affect the subject's rights, safety and well-being, or the scientific integrity of the clinical investigation.

Any deviation from the protocol that has not been previously approved by the Principal investigators must be reported to the Central contact person within 3 working days of the deviation occurrence. Any deviations from the clinical investigation plan that are identified during routine monitoring visits will be reported to the Central contact person within 3 working days of being identified.

If possible, prior approval from the Primary investigators, if appropriate, shall be obtained by the investigator. All spontaneous CIP deviations shall be recorded and reported to the central Contact person as agreed. A deviation log shall be maintained by the study site. Deviations shall be reported to the regulatory authorities if required by national regulations. All deviations will be included, as required in the final study report.

Participants whose parents have consented to intervention, assessments, follow-up and data collection, their participation is voluntary and they are free to withdraw at any time and without their medical care or legal rights being affected. A participant may be withdrawn from trial whenever continued participation is no longer in the participant's best interests, and the reason for withdrawal will be recorded. Reasons for discontinuing the trial may include:

- intercurrent illness
- patients withdrawing consent
- persistent non-compliance to protocol requirements

The decision to withdraw a participant from treatment will be recorded in the CRF and medical notes.

5.5 Quality control: monitoring plan

A site-specific monitoring plan will be assigned to follow the quality and justification of the methods used, and fulfilment of the study patients' rights and their well-being.

The Primary investigators will determine the appropriate level and nature of monitoring required for the trial. Risk will be assessed on an ongoing basis and adjustments made accordingly. The degree of monitoring will be proportionate to the risks associated with the trial. A trial specific oversight and monitoring plan will be established for studies. The trial will be monitored in accordance with the agreed plan.
Low risk: central monitoring. Each site to email to the Central contact person annually: delegation log, adverse event log, deviation log, minutes of local Ethics board (or equivalent).

We propose that no interim analysis is planned, unless excess of adverse effects are observed.

6 Pharmacokinetics

The paracetamol serum levels will be measured from all the study patients in study sites where this is possible to accomplish. All the routine laboratory samples' left-overs will be recovered during the study drug period. These will be used for measurements that include serum or plasma paracetamol

concentrations, and possibly paracetamol metabolites. The exact administration dates and time points of the study drug will be recorded, as well as dates and time points of the blood samples collected.

As soon as blood is collected, they will be stored for future research. An appropriate SOP will take place in each site. The specimen will be posted to the coordinating centre, where they will be stored at -70 degrees C in the Research laboratory of Department of Paediatrics, University of Oulu.

7 Data management procedures

When the parents agree to participate, demographic, contact and medical history information necessary to conduct the study will be recorded in accordance with the patient consent form, patient information sheet and section 4 of this protocol. Each participant will be allocated a unique trial number. This data will be collected on a CRF, on a secure server; sealed envelope. Access to this data, will be granted to authorized research staff, host institution and regulatory authorities to permit trial-related activities, monitoring and audit. The study registry has been assigned to the Finnish Data Protection Ombudsman's Office.

All the trial data is confidential. The whole study personnel are bound to secrecy. The data will be collected to specified files stored into the hospital datasets of the Primary investigator's affiliation.

Double-checking of the input of individual data for errors is imperative. Data clarification will be issued to the investigator should a discrepancy be found between the source and CRF. The investigator will be required to verify and correct all errors or provide an explanation for the discrepant data.

All data from the examinations and investigations listed in will be transferred to coordinating centre by posting or, if available, by secured email system. The Central contact person will manage and maintain the study database throughout the investigation. At the conclusion of the investigation, the database will then be locked and data transferred for analysis. There will be a documented record of data transfer in place for the recovery of original information after transfer. The database maintained by the Central contact person shall be validated and secured according to the coordinating centre's (Oulu University Hospital) standard operating procedures. Access to the data shall be limited to investigators directly involved in the collection, analysis, maintenance or safety monitoring of the data, and regulatory authorities.

Any study data released shall be done according to the publication policy. The study results will be published in high-quality international medical series and theses.

7.1 Trial data in CRF

From all study infants, case report files (annex 2) will be filled out. It contains following information:

- Gestational parameters: maternal age and diagnoses, parity, medications during the pregnancy, possible complications, the amount of amnion fluid, cause of premature birth
- Infants: gestational age, gender, birth measurements (weight, length, head circumference), PDA therapies, pain medication doses, diuresis.
- Studies: cerebral and abdominal ultrasound scan reports, thorax x-ray findings
- The timeline of the study drug course: given doses, blood samples taken, diapers weighted, ongoing NIRS recordings, blood pressures measured, ultrasound examinations
- Laboratory values: transaminase, bilirubin and cystatin-C values, if taken
- Diagnoses: PDA, BPD, ICH, NEC, ROP

7.2 Statistical analysis plan

In general, the most suitable statistical analysis consistent with the form of data will be used and the underlying assumptions of the statistical method will be verified. All trial data analyses will be performed according to the intention-to-treat principles. All tests will be two-sided; a p value <0.05 will be considered to indicate statistical significance.

Gestational age at birth, postnatal age (SD), sex, birth weight, and other medical history will be used for baseline comparability of the randomised groups.

The primary outcome, the difference in ductal closure rate (and the difference in change of repeated PDA therapies) between randomised groups will be compared using t-tests. The secondary outcomes will be analysed using t-test, Chi squared test, and regression analyses depending on the variables.

SPSS software, version 23.0.0.0 (IBM SPSS Statistics, NY, USA) will be used to conduct analyses.

8 Project management

The principal investigators, investigators, and the site staff are qualified for the management and execution of the trial. The trial will take place in high-quality, tertiary care university hospitals. The study locations will be their neonatal intensive care units that provide the most advanced and specialised standards for care and appropriate, professional equipment for the treatment of the premature infants with adequate preparation for the emergencies as well. All the trial sites are considered suitable for this trial.

8.1 Permissions and practices

The following permissions for the original one-centre trial have been given:

- TUKIJA
- The attending hospital's regional ethics board and the administration of the department
- Finnish Medical Agency (Fimea)

The substantial amendments for the multicentre trial permissions will be applied from both organizations. Necessary permissions from the corresponding organizations of the other Finnish sites (Helsinki, Turku) and Estonia will be applied as well.

Present trial has been assigned in the following registries:

- European Clinical Trials Database, ID 2018–000565-36
- ClinicalTrials.gov registry, ID NCT03648437

The present trial is and will be conducted by applying the principles of Good Clinical Practice (Fimea directive II/2012, EMA Guideline for good clinical practice E6[R2]). The essential changes in the investigation plan will be announced to Fimea according to the Drug act, moment 87a. This will be announced using the European Commission internet form (http://ec.europa.eu/health/documents/eudralex/vol-10/).

8.2 Amendments to the CIP

Amendments to this CIP may be necessary to protect the safety of the patients and integrity of the data. In collaboration with the Primary investigators, the CIP amendments will be documented and submitted for ethical and regulatory approval (as required) prior to implementation. All changes will be evaluated for impact. Amendments will be considered implemented after all ethical and regulatory approvals (as required) are received and all key site staff has been trained. This process does not affect the individual clinician's responsibility to take immediate action if thought necessary to protect the health and interest of individual patients.

8.3 Timetable estimation of the multicentre trial

2018 - 2019: permissions
2018 - 2022: recruitment of the patients
2022 - 2023: data analysis and reporting.

8.4 Long-term follow-up

According to the recommendations of the Cochrane database report, the long-term follow-up trials are planned for the trial patients.[12] They will be accomplished at the 2 to 3 years' age and/or at 5 to 7 years' age of the study patients. However, these trials will be managed and executed by the separate research plans and the permissions of their own will be applied.

9 Ethical considerations

The randomised, controlled, double-blind study design is the only possibility to objectively evaluate the efficacy and safety of the drug. It is ethically imperative that all new indications should be tested within the patient population in question. This is the goal of the European Union drug directive on paediatric patients as well. It would be highly unethical to implement a new indication without acquiring relevant evidence.

According to previous studies in preterm infants, the chosen dose of the study drug is not a risk in preterm infants as no side effects, or any harmful long-term effects, have been found in the paracetamol group infants compared to the controls.

The study laboratory tests are well tolerated for the smallest preterm infants as well. The pain and discomfort of the study infants are being treated during the intervention. Local patient pain screening scoring systems will be used for observing the need for pain therapy in preterm infants. These scales will be used in the present study by trained and experienced nurses. The aim is to keep the infant painless and calm despite the intensive care. The standard medication (e.g. intravenous morphine) has been shown to be effective on the pain therapy of preterm infants.

The participation in the study is completely voluntary. If the screened patient will not participate in the study, it would not have any consequences on any treatments during the hospital stay. All parents are asked the informed written consent. We consider the long-term follow-up very important as well, and hope to be able to accomplish it.

10 Annexure

1. Informed consent forms: Finnish and Estonian
2. Case record form (CRF)

11 References

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