Date: 14th July, 2022

Study Protocol

Project title: Randomized controlled trial comparing the efficacy and safety of mydriatic microdrops over standard dose mydriatics for pupil dilation in retinopathy of prematurity examination.

Background and purpose:
Preterm and very low birth weight infants are at risk of developing retinopathy of prematurity (ROP) hence they require regular screening for early detection and timely treatment for ROP. Mydriatics are applied before ROP exam to dilate the pupils for fundal examination. In our unit, we are using Mydrin-P (0.5% tropicamide / 0.5% phenylephrine HCl) given thrice at 15 minutes intervals. This total dosage is the same as is being used for adult patients. Mydriatics are well known to have systemic side effects such as hypertension or hypotension, tachycardia or bradycardia, apnoea, cardiopulmonary arrest, seizures, necrotizing enterocolitis, and sepsis as they can bypass hepatic first pass metabolism and be directly absorbed into the systemic circulation. Preterm infants are more prone to suffer from these side effects due to smaller blood volume and immature drug metabolism. [10 13 14] The smallest efficacious dose and combination of mydriatics for use in preterm infants has yet to be documented.

Reduction in mydriatics dose for ROP exam emerges as a promising method to minimize systemic side effect without compromising the efficacy of mydriatics. [1 2 3] Several pilot studies in non-Asian countries were able to show that lower doses of mydriatics can achieve adequate pupil dilation [16 17]. A systemic review evaluated 47 publications from 1986 to 2017 on different mydriatics preparation used for ROP exam. It was shown that the lower doses mydriatics are effective compared to current high dose mydriatics being used following international guidelines. [14] However, most published data are underpowered and the population group in those studies are mostly non-Asians. It is known that that darker irides in Asians or Africans are more difficult and slower to dilate.[20] The study on Asians by Luo et al. mentioned in the systemic review only compared the commercial Mydrin-P over locally compounded eye drops with the same concentration. [14, 19] There have been no studies or trials attempting to reduce the dose of Mydrin-P for use in ROP eye exams. Hence, we hope to be the pioneer in this area to conduct a non-inferiority trial comparing the efficacy and safety of microdrops over standard dose Mydrin-P in ROP screening.

Aim of the study:
To investigate

1. If microdrops Mydrin-P can be used in ROP exam with similar efficacy compared to our current standard dose Mydrin-P, potentially with the advantage of fewer adverse effects.
2. To ascertain the optimal time for eye examination after administration of mydriatics.
3. To assess whether the cardiovascular, respiratory and gastrointestinal adverse effects differ between low dose mydriatics compared to standard dose.

Inclusion and exclusion criteria:
All preterm infants eligible for ROP screening will be recruited including those with estimated gestational age (EGA) at birth ≤32 weeks and/or birth weight ≤1500g. Neonates

with severe clinical condition with unstable vital signs, congenital anomalies, syndromic disease, ophthalmological conditions such as eye infections, congenital eye anomalies, trauma, or with conditions that are contraindication to mydriatics will be excluded from the trial.

Methods:

This is a prospective, single centre, double blinded randomized controlled trial to be conducted on a group of preterm infants undergoing retinopathy of prematurity (ROP) examination. The study will be conducted from around third quarter of 2022 (July-September) to end of 2023. Inpatient infants will be recruited from Queen Mary Hospital in the neonatal intensive care unit and special baby care unit. Informed consent will be obtained from parents. Participants will be randomly assigned to either the study group or control group through computer generated algorithm for each episode of ROP exam. Randomization ratio is assigned to be 1:1. Each patient may have more than 1 episode of ROP examination so each episode of ROP exam will be randomly allocated into study or control group. The randomization result will be concealed to principal investigator and ophthalmologist who will conduct the ROP exam. The control group will receive the standard Mydrin-P eyedrops which contain 0.5% phenylephrine HCl and 0.5% tropicamide. The study group will receive mydriatic microdrops which contain around one-third of the standard Mydrin-P dosage.

The demographics including sex, gestational age and corrected age, current weight, ethnicity, level of respiratory support, eye color, other comorbidities such as bronchopulmonary dysplasia, necrotizing enterocolitis, renal failure, patent ductus arteriosus, sepsis etc. will be obtained prior to ROP examination.

Following the randomization, those allocated to the control group will receive standard Mydrin P eyedrops (0.5% tropicamide / 0.5% phenylephrine HCl). Those allocated to the study group will receive Mydrin-P microdrops. The microdrop administration is through attaching a 16 gauge needle to a 1ml syringe. Drop volume measurement was performed. A precision weight scale (Sartorius) with accuracy up to 0.001g was used to measure the drop volume. Standard Mydrin-P volume is around 45microlitre. Microdrop volume is around 16microlitre which is around one-third of the standard Mydrin-P drop size.

The eyedrops will be applied to the patients by nurses through a standardized method. All nurses will be taught on the methods of eyedrop application in order to reduce variations. Each dose of eyedrop should be applied from the medial corner of the eye directed to the lateral corner. Drug spilling around the eyes will be wiped off. Then a slight pressure should be applied to the medial corner of the eye for 3 minutes to prevent nasal mucosal absorption. For both groups of babies, one drop will be administered every 15 minutes for three administrations in total as per current practice prior to ROP screening by the ophthalmologist.

Sample size calculation

The sample size calculation was based on mean and standard deviation. Referring to the study published by Luo et al.[19], mean pupil diameter was 7.22mm with 0.4mm standard deviation. The sample size was calculated based on the margin of error of this effect. With a

sample size of 30, 95% of the confidence interval will be from 7.067 to 7.36mm with a margin of error of 0.15mm. Moreover, our pre-set cut off for minimal pupil diameter adequate for ROP exam is 7mm which was agreed by our ophthalmologist, with this sample size, it is estimated that 95% of the subject will have greater than or equal to a pupil size of 7.067mm for ROP exam.

Statistical analysis:
IBM SPSS statistics version 26 software will be used for all statistical analysis. Demographic data are expressed in mean, standard deviation, number, and percentage when appropriate.

Outcome including mean pupil diameter, vital signs such as heart rate, blood pressure, oxygen saturation, FiO2 requirement, episode of vomiting, volume of gastric residuals, episode of apnoea and the presence of periorbital blanching will be measured and compared between the study and the control group. Fisher's exact test will be used for categorical variables, independent samples T test will be used for continuous data. A paired sample t-test will be used to compare the related two means between paired observations. Significance level was set at 5%. All ethical considerations will be ensured throughout the study.

Study outcomes:
A trained masked observer will assess pupil size by using a pupillometer and visual assessment to capture 2 readings from each eye for each patient at four timepoints. Pupil diameter (in millimetres, mm) will be documented at baseline (before eyedrops are applied, T0), 30 minutes later (T30), 60 minutes later (T60) and 120 minutes later (T120). An eye speculum may need to be inserted by a registered trained doctor to open up the eyelids during pupillometer measurements to obtain the readings if spontaneous eye opening is not possible. Ophthalmologist will perform the ROP exam after the 3 doses of mydriatics. If there is inadequate pupil dilation as determined by the ophthalmologist, additional doses of standard dose Mydrin-P will be given in order to facilitate completion of the exam. And these patients would be considered as failure for exam and the data will not be included in the final analysis. Time of fundoscopy, the number of additional doses of mydriatics given will be documented.

The primary outcome is whether the pupil size has attained equal or more than 7mm for ROP exam. 7mm will be the cutoff we use for an adequate pupil diameter for ROP exam.

The secondary outcomes are the pupil diameters at T0, T30, T60 and T120, cardiovascular, respiratory, gastrointestinal side effects followed by the mydriatics administration. Vital signs such as heart rate, blood pressure (systolic and diastolic blood pressure taken at the same body part), oxygen saturation, FiO2 requirement will be documented at baseline (T0, just before eyedrop application), 30 minutes later (T30), 60 minutes later (T60) and 120 minutes (T120). The following data will also be documented in the 24 hours after eyedrops administration and compared to the baseline which is the past 24 hours prior to the eyedrops exposure: episodes of vomiting, volume of gastric residuals, episodes of apnoea and the presence of periorbital blanching. Any other adverse event will also be recorded.

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