

Comparison of microdrop administration using a bottle adapter of 2.5% phenylephrine and 0.8% tropicamide with standard drop for ROP examination: a randomized controlled noninferiority trial

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Abstract

Background

Pharmacologic mydriasis is essential for retinopathy of prematurity (ROP) screening; however, conventional ophthalmic drops substantially exceed neonatal tear film capacity, leading to systemic absorption and potential adverse effects. Microdrop administration may reduce drug exposure while preserving adequate pupillary dilation. This study aimed to determine whether microdrop administration of phenylephrine and tropicamide is noninferior to standard drops in achieving adequate mydriasis for ROP screening via a bottle adapter.

Methods

This single-center, randomized, parallel-group noninferiority trial was conducted in a tertiary neonatal unit in Chennai, India. Preterm infants undergoing routine ROP screening were randomized to receive either microdrops (10.26 μ L) or standard drops (26.58 μ L) of 2.5% phenylephrine and 0.8% tropicamide. Three doses were administered at 10-minute intervals. The primary outcome was the mean pupil diameter at 45 minutes after the first instillation. The secondary outcomes included physiological parameters (heart rate, oxygen saturation, and blood pressure), systemic and local adverse events, and the adequacy of ROP examination. A noninferiority margin of -0.5 mm was prespecified.

Results

Among the 127 infants assessed for eligibility, 102 were randomized (microdrop group, $n = 52$; standard group, $n = 50$), and all completed the study. Baseline characteristics were comparable between the groups. The mean pupil diameter at 45 minutes was 6.54 ± 0.52 mm in the microdrop group and 6.38 ± 0.63 mm in the standard group (mean difference, 0.16 mm; 95% CI -0.07 – 0.38), demonstrating noninferiority. Physiological parameters, including heart rate, oxygen saturation, and blood pressure, remained stable and comparable between the groups. Systemic adverse events were rare and similar across groups, and no local ocular complications were observed. ROP examination was successfully completed in all infants.

Conclusion

Microdrop administration of phenylephrine and tropicamide achieved mydriasis comparable to that of standard drops without increased adverse effects. This approach may represent a safer, clinically feasible and more physiologically appropriate strategy for ROP screening by reducing drug exposure in vulnerable preterm infants.

Trial registration

The study protocol was approved by the Institutional Ethics Committee of Madras Medical College, Chennai (IEC No. MMC/Approval/12052025). The trial was registered with the Clinical Trials Registry of India (CTRI/REF/2025/03/101681) on 10-03-2025.

Introduction

Preterm neonates undergo multiple examinations for retinopathy of prematurity (ROP) during their stay in the neonatal intensive care unit (NICU) and after discharge for early diagnosis and timely management. Various mydriatic regimens with different compositions are used worldwide. The commonly used topical agents include a combination of adrenergic agonist and anticholinergic agents. Up to 80% of instilled ophthalmic drops may undergo systemic absorption, particularly in neonates, due to a reduced tear film volume and immature drug metabolism[1, 2]. Standard ophthalmic drops (~ 26 µL) substantially exceed neonatal tear film capacity (0.5–2 µL), resulting in overflow and nasolacrimal drainage[3]. In contrast, microdrops (5–15 µL) better approximate the physiological tear film volume and may reduce systemic exposure while maintaining efficacy[4–6].

Several studies have demonstrated that reduced-volume mydriatic administration via micropipettes and modified cannulas achieves comparable pupillary dilation with reduced systemic exposure[4–8].

Currently, there are no studies evaluating the mydriatic regimen commonly used in India for neonatal ROP screening with a simplified delivery method, such as bottle adapters or calibrated droppers, which could facilitate the widespread use of microdrop instillation across neonatal units. The present study aimed to evaluate the efficacy and safety of microdrops (10.26 µL) compared with standard drops (26.58 µL) of the recommended formulation containing 2.5% phenylephrine and 0.8% tropicamide in achieving adequate mydriasis for ROP examination.

Methods

Primary Objective

To determine whether microdrop administration of 2.5% phenylephrine and 0.8% tropicamide is noninferior to standard drop administration in achieving adequate pupillary dilation 45 minutes after the first instillation in preterm infants undergoing ROP screening.

Secondary objectives

- To assess the incidence of local adverse effects following mydriatic administration
- To evaluate systemic physiological changes, including heart rate, oxygen saturation, and blood pressure

- To assess the adequacy of ROP examination and the ability to identify treatment-requiring ROP

Study Design

This study is a single-center, two-arm, parallel-group, randomized noninferiority trial.

Study Setting

The study will be conducted in the Department of Neonatology, Institute of Child Health, Madras Medical College, Chennai, India.

Study population

Preterm neonates undergoing routine ROP screening according to institutional and national guidelines will be eligible for participation.

Study Period

The study was conducted over a period of **six months** from April 2025 to October 2025.

Eligibility criteria

Inclusion criteria

- Preterm infants with a **gestational age < 34 weeks** or **birth weight < 2 kg**
- Infants with a gestational age of **34–36 weeks** with additional risk factors, including the following:
 - Requirement for cardiorespiratory support
 - Prolonged oxygen therapy > 24 hours
 - Respiratory distress syndrome
 - Bronchopulmonary dysplasia
 - Neonatal sepsis (culture positive)
 - Blood transfusion
 - Intraventricular hemorrhage
 - Apnea
 - Poor postnatal weight gain (< 15 g/kg/day)

Exclusion criteria

- Unstable vital signs
- Requirement of inotropes, phenobarbitone, benzodiazepines, opioids, ketamine, and anticholinergics in the last 48 hours.
- Corneal pathology or anterior segment anomalies
- ROP stage > 2 at presentation

Sample size

On the basis of a power of 90%, an assumed standard deviation of 0.75 mm (based on a study by alpey et al.), and a predefined noninferiority margin of – 0.5 mm for the pupillary diameter, a minimum sample size of 78 infants is needed[9].

Considering a potential 20% dropout rate, the total sample size will be 98 infants, with 49 infants in each group.

The noninferiority margin of 0.5 mm was determined on the basis of clinical consensus as the largest difference that would not meaningfully affect the adequacy of mydriasis needed for ROP screening. This margin was also pragmatic, as pupillary size was measured via a customized ruler marked at 0.5-mm intervals. Prior trials evaluating microdrop mydriasis have used similar margins, supporting the clinical relevance of this range[1, 7, 8]. Setting the noninferiority margin at this unit mitigates measurement error and ensures alignment between the statistical sensitivity and the practical limits of clinical precision.

Randomization and Blinding

- Eligible infants were identified by the treating clinician and enrolled after eligibility was confirmed.
- Participants underwent stratified randomization on the basis of gestational age (< 34 weeks and ≥ 34 weeks), and block randomization with block sizes of 2, 4 and 6 was performed within each stratum. The random allocation sequence was computer generated via the Sealed Envelope online randomization tool (Sealed Envelope Ltd., London, UK).
- Allocation concealment was ensured via sequentially numbered, opaque, sealed envelopes prepared by an independent statistician.
- The nursing staff responsible for administering the eye drops were aware of the group allocation due to the nature of the intervention.
- The pupillary diameter (in millimeters) was measured by the outcome assessor via a customized ruler with 0.5-mm incremental markings.
- The outcome assessor, investigators, and data analysts remained blinded to group allocation throughout the study.

Intervention

Following randomization, eligible neonates were allocated to one of the two study groups:

Microdrop group (intervention):

A microdrop volume of **10.26 µL** was administered via a sterile bottle adapter (**Nanodropper**), as shown in Fig. 1.

Standard drop group (comparator):

A standard drop volume of **26.58 µL** was instilled directly from a conventional synthetic dropper bottle.

The **same mydriatic formulation** will be used in both groups:

- Phenylephrine 2.5% and tropicamide 0.8% (TrophthaTM-semi)

In both groups, three doses will be administered by the nurse, with one drop instilled into each eye at 10-minute intervals.

During administration, the infant's eyelids were gently retracted by the nurse, and one drop was instilled into each eye at each dosing interval.

During the procedure, infants were swaddled and continuously monitored for vital parameters via a multiparameter monitor. An ROP examination was performed by an experienced ophthalmologist via the RetCam imaging system.

Prior to examination, topical prilocaine anesthetic eye drops were instilled, after which an eyelid speculum was gently inserted. Retinal examination and staging were performed and documented via RetCam images.

Outcome assessment

Primary outcome

The primary aim of this study was to determine whether microdrop administration of 2.5% phenylephrine and 0.8% tropicamide is noninferior to standard drop administration of the same mydriatic regimen in achieving adequate pupillary dilation 45 minutes after the first drop instillation (T45) in preterm infants undergoing ROP screening.

Secondary outcome

- To evaluate heart rate (HR), oxygen saturation (SpO₂), systolic (SBP), diastolic (DBP), and mean (MBP) blood pressure at T45, T90, and T120 [time frame: 45 minutes, 90 minutes, 120 minutes], respectively,
- Number of participants with systemic adverse events. [Time frame: During the 48 hours after mydriasis] - Apnea, increased gastric residuals, feeding intolerance, abdominal distension, vomiting, NEC.

- Number of participants with local adverse events. [Time frame: 45 minutes after the first drop instillation] - Periorbital pallor, eyelid swelling, flushing
- Adequacy of determining the presence or absence of treatment-requiring ROP. [Time frame: At the end of the eye examination (fundoscopy)]

Baseline Measurements

At baseline, the following parameters were recorded prior to mydriatic administration:

- Pupillary diameter
- Heart rate (HR)
- Oxygen saturation (SpO₂)
- Systolic blood pressure (SBP)
- Diastolic blood pressure (DBP)
- Mean blood pressure (MBP)

HR and SpO₂ were measured via a Massimo monitor, and BP was measured via a Nikon koiden monitor.

Assessment of Mydriatic Efficiency

Pupillary dilation was assessed under bright illumination using a pen torch. The eyelids were gently opened, and the pupillary diameter was measured via a customized ruler with 0.5-mm incremental markings, as shown in Fig. 2.

Measurements were obtained for both eyes, and the lower measurement among the two eyes was considered a single value for analysis.

Timing of measurements

Pupillary diameter and physiological parameters were recorded at the following time points:

- **T45:** 45 minutes after the first drop instillation
- **T90:** 90 minutes after the first drop instillation
- **T120:** 120 minutes after the first drop instillation

Secondary outcome definitions

The secondary outcomes included the occurrence of systemic and gastrointestinal adverse events following mydriatic administration. The following operational definitions were used:

Apnea

Cessation of breathing for ≥ 20 seconds or a shorter respiratory pause associated with bradycardia (heart rate < 100 beats/min) and/or oxygen desaturation[10].

Increased Gastric Residuals

Aspiration of gastric contents exceeding 2 mL/kg or > 50% of the previous feed volume following enteral feeding, assessed in preterm infants receiving feeds via an orogastric or nasogastric tube[11].

Feeding intolerance

The inability to tolerate enteral feeds is characterized by increased gastric residual, abdominal distension, and/or vomiting[11].

Abdominal Distension

A measurable increase in abdominal girth of ≥ 2 cm compared with baseline[11].

Necrotizing enterocolitis (NEC)

is defined as Bell's stage II or higher[12].

Hypertension

Blood pressure greater than the 95th percentile for gestational and postnatal age, according to the Zubrow neonatal blood pressure reference chart[13, 14].

Statistical analysis

Statistical analysis was performed via SPSS version 25 (IBM Corp., Armonk, NY, USA). All randomized infants who received the assigned intervention and had baseline measurements were included in the analysis.

Continuous variables (gestational age, postmenstrual age, birth weight, present weight, pupil diameter, heart rate, SpO₂, systolic blood pressure, diastolic blood pressure, and mean blood pressure) are expressed as the mean (SD) or median (interquartile range), as appropriate, and categorical variables (sex, comorbidities, ROP stage, adverse events, respiratory support, and examination adequacy) are expressed as frequencies and percentages. The normality of the distribution was assessed via the Shapiro–Wilk test. Baseline characteristics were compared between groups via the independent samples t test for continuous variables and the chi-square test or Fisher's exact test for categorical variables.

The primary outcome was the mean pupil diameter at 45 minutes (T45). The noninferiority of microdrops compared with standard drops was evaluated via the mean difference (microdrop – standard) and the corresponding two-sided 95% confidence interval (CI). A noninferiority margin of – 0.5 mm was predefined. Noninferiority was concluded if the lower bound of the 95% CI was above the prespecified margin[15–17].

Between-group comparisons of pupil diameter at T45, T90, T120, and postprocedure were performed via independent samples t tests. Within-group changes from baseline were analyzed via paired t tests.

Physiological parameters (heart rate, SpO₂, and systolic, diastolic, and mean blood pressure) at baseline and subsequent time points were compared between groups via independent samples t tests.

The incidence of systemic and local adverse events and the adequacy of ROP examination were compared via the chi-square test or Fisher's exact test, as appropriate.

A predefined subgroup analysis was performed according to gestational age (≤ 33 weeks and > 33 weeks). All tests were two-sided, and a p value < 0.05 was considered to indicate statistical significance. No missing data or protocol deviations were observed; therefore, no imputation or sensitivity analyses were performed.

Results

A total of 127 infants were assessed for eligibility, 102 of whom were randomized: 52 to the microdrop group and 50 to the standard drop group. Twenty-five infants were excluded (18 did not meet the inclusion criteria, 5 declined participation, and 2 were unable to provide consent). All randomized participants received the allocated intervention and were included in the final analysis, with no loss to follow-up. The CONSORT flow diagram of participant recruitment, randomization, analysis and follow-up is shown in Fig. 3.

Baseline characteristics

The baseline demographic and clinical characteristics were well balanced between the groups, with no statistically significant differences. The mean (SD) gestational age was 30.9 (3.2) weeks in the microdrop group and 30.7 (3.3) weeks in the standard group. The mean (SD) birth weights were 1401 (563) g and 1327 (606) g, respectively. The distributions of retinopathy during prematurity stages and comorbidities, including bronchopulmonary dysplasia, patent ductus arteriosus, necrotizing enterocolitis, and sepsis, were comparable between the groups. The baseline demographic and clinical characteristics are summarized in Table 1, Table 1 and Table 2.

Table 1
Baseline characteristics

Variable	Micro drops (n = 52)	Standard drops (n = 50)	P value
Gender	31(59.6%)	23(46.0%)	0.168 ^{\$}
Male	21(40.4%)	27(54.0%)	
Female			
Post menstrual age (in weeks)	36.67(3.58) 26–44	36.32(3.43) 30–44	0.613*
Mean(SD)			
Min – Max			
Gestational age (in weeks)	30.85(3.19)	30.72(3.32)	0.845*
Mean(SD)	25–36	25–36	
Min – Max			
Birth weight (in gms)	1400.79(563.40)	1326.54(606.39)	0.523*
Mean(SD)	580–3200	580–3600	
Min – Max			
Present weight (in Kgs)	1560.90(637.72)	1543.50(685.11)	0.895*
Mean(SD)	610–3500	610–3700	
Min – Max			
ROP Stage (Previous exam)	29(55.8%)	25(50.0%)	0.590 ^{\$}
0	12(23.1%)	10(20.0%)	
1	11(21.1%)	15(30.0%)	
2			
Sepsis	30(57.7%)	33(60.0%)	0.388 ^{\$}
BPD	7(13.5%)	2(4.0%)	0.092 ^{\$}
PDA	12(23.1%)	14(28.0%)	0.568 ^{\$}
NEC	8(15.4%)	5(10.0%)	0.415 ^{\$}
^{\$} Chi-square test/Fisher's exact test; *t test			

Variable	Micro drops (n = 52)	Standard drops (n = 50)	P value
Feed intolerance	2(3.8%)	2(4.0%)	0.968 ^{\$}
Vomiting last 24 hour	1(1.9%)	2(4.0%)	0.614 ^{\$}
Increased feed residuals	1(1.9%)	-	0.999 ^{\$}
Abdominal distension	1(1.9%)	1(2.0%)	0.999 ^{\$}
Respiratory support	1(1.9%)	-	0.566 ^{\$}
MV	5(9.6%)	5(10.0%)	
NIV	40(76.9%)	35(70.0%)	
RA	6(11.5%)	10(20.0%)	
CPAP			
Apnea episodes past 24 hr	8(15.4%)	10(20.0%)	0.541
Inotropes	15(28.8%)	17(34.0%)	0.575 ^{\$}
Post natal steroids	-	1(2.0%)	0.490 ^{\$}
^{\$} Chi-square test/Fisher's exact test; *t test			

Table 2

Pupillary diameter at baseline and after mydriatic instillation (primary outcome at 45 minutes)

Pupil Diameter	Micro drops (n = 52)	Standard drops (n = 50)	Diff	95% CI for diff	Diff-Margin	SE	T	P value*
Baseline	1.59(0.40)	1.68(0.40)	-0.09	-0.25 to 0.06	0.41	0.080	5.12	< 0.001
Mean(SD)	1–2	1–2						
Min – Max								
Time 45(primary outcome)	6.54(0.52)	6.38(0.63)	0.16	-0.07 to 0.38	0.66	0.114	5.79	< 0.001
Mean(SD)	5–7.5	5–7.5						
Min – Max								
Time 90	6.60(0.56)	6.42(0.71)	0.18	-0.07 to 0.43	0.68	0.126	5.40	< 0.001
Mean(SD)	5–7.5	4.5–7.5						
Min – Max								
Time 120	6.39(0.64)	6.25(0.74)	0.14	-0.13 to 0.42	0.64	0.138	4.67	< 0.001
Mean(SD)	4.5–7.5	4–7.5						
Min – Max								
Post procedure	6.58(0.54)	6.40(0.73)	0.18	-0.075 to 0.43	0.68	0.127	5.35	< 0.001
Mean(SD)	5–7.5	4–7.5						
Min – Max								
*t test								

Primary outcome

At 45 minutes, the mean (SD) pupil diameter was 6.54 (0.52) mm in the microdrop group and 6.38 (0.63) mm in the standard group, with a mean difference of 0.16 mm (95% CI, – 0.07–0.38), meeting the prespecified criterion for noninferiority. Pupillary diameter measurements at baseline and following mydriatic instillation are presented in Table 2. The results of the forest plot analysis of the primary outcome and gestational age subgroup analysis are shown in Fig. 4.

Subgroup analyses demonstrated consistent findings across gestational age strata. Among infants younger than 34 weeks, the mean difference was 0.13 mm (95% CI, – 0.11–0.45), and among those 34

weeks or older, the mean difference was 0.10 mm (95% CI, - 0.28–0.49). The results of the subgroup analysis of the primary outcome are shown in additional Tables 3 and 4.

Table 3
Heart rate measurements at each time point

Heart rate	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Baseline	139.56(8.53)	138.36(8.65)	0.469
Mean(SD)	120–164	125–160	
Min – Max			
45 minutes	139.83(8.42)	140.10(9.11)	0.875
Mean(SD)	118–162	125–168	
Min – Max			
90 minutes	139.44(8.85)	139.60(8.54)	0.927
Mean(SD)	120–166	125–158	
Min – Max			
120 minutes	139.36(9.08)	138.22(7.96)	0.500
Mean(SD)	118–162	125–156	
Min – Max			
*t test			

Table 4
SPO₂ measurements at each time point

SPO₂	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Baseline	96.60(1.78)	96.36(1.91)	0.521
Mean(SD)	92–99	91–99	
Min – Max			
45 minutes	97.02(1.58)	96.56(1.73)	0.164
Mean(SD)	92–99	92–100	
Min – Max			
90 minutes	97.04(1.55)	96.54(1.90)	0.148
Mean(SD)	93–99	90–99	
Min – Max			
120 minutes	97.02(1.54)	96.78(1.45)	0.421
Mean(SD)	93–99	92–99	
Min – Max			
*t test			

Secondary Outcomes

There were no significant differences in the physiological parameters between the groups at any time point. The mean heart rate, oxygen saturation, and systolic [SBP], diastolic [DBP], and mean arterial blood pressure (MBP) remained comparable at 45, 90, and 120 minutes following drug administration.. Heart rate measurements at each time point are shown in Table 3, SpO₂ measurements are shown in Table 4, and blood pressure measurements are shown in Table 5.

Table 5
Blood pressure measurements at each time point

SBP	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Baseline	71.90(7.37)	69.98(9.55)	0.256
Mean(SD)	51–86	56–120	
Min – Max			
45 minutes	71.48(7.62)	70.36(11.10)	0.552
Mean(SD)	51–86	52–126	
Min – Max			
90 minutes	71.58(6.93)	70.44(10.32)	0.514
Mean(SD)	59–84	55–125	
Min – Max			
120 minutes	71.56(7.66)	70.14(10.21)	0.428
Mean(SD)	51–84	52–120	
Min – Max			
DBP	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Baseline	39.33(6.88)	38.92(7.49)	0.776
Mean(SD)	25–54	27–56	
Min – Max			
45 minutes	40.79(6.18)	39.68(7.62)	0.421
Mean(SD)	30–55	22–60	
Min – Max			
90 minutes	40.44(6.79)	39.82(6.70)	0.643
Mean(SD)	30–54	30–55	
Min – Max			
120 minutes	41.56(6.68)	40.12(6.82)	0.285
Mean(SD)	32–56	27–56	
*t test			

SBP	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Min – Max			
MBP	Micro drops (n = 52)	Standard drops (n = 50)	P value*
Baseline	49.49(6.37)	49.28(7.11)	0.876
Mean(SD)	33–62	37–72	
Min – Max			
45 minutes	50.61(6.16)	48.90(7.30)	0.202
Mean(SD)	38–63	32–70	
Min – Max			
90 minutes	49.94(6.49)	49.52(6.79)	0.749
Mean(SD)	39–62	36–70	
Min – Max			
120 minutes	51.06(6.54)	49.58(6.60)	0.259
Mean(SD)	39–63	37–72	
Min – Max			
*t test			

For example, at 45 minutes, the mean (SD) heart rate was 139.8 (8.4) beats/min in the microdrop group and 140.1 (9.1) beats/min in the standard group (mean difference, – 0.27; 95% CI, – 3.5–3.0; $P = .88$). Similar nonsignificant differences were observed for oxygen saturation and blood pressure parameters across all measured time points.

Safety and Clinical Outcomes

There were no significant differences in adverse events between the groups. The incidences of apnea, gastrointestinal symptoms (including vomiting, abdominal distension, and NEC), and the need for respiratory or inotropic support were comparable. The postprocedure clinical outcomes and adverse events are summarized in Table 6.

Table 6
Postprocedure clinical outcomes and adverse events

Post procedure	Micro drops (n = 52)	Standard drops (n = 50)	P value
Local reactions	-	-	-
Abdominal distension	-	1(2.0%)	0.490
Vomiting	1(1.9%)	1(2.0%)	0.999
Increased gastric	-	-	-
NEC	-	-	-
Apnea	9(17.3%)	12(24.0%)	0.403
Respiratory support	1(1.9%)	-	0.139
MV	5(9.6%)	5(10.0%)	
NIV	42(80.8%)	34(68.0%)	
RA	4(7.7%)	11(22.0%)	
CPAP			
Successful ROP exam	52(100.0%)	50(100.0%)	-
ROP stage	21(40.4%)	20(40.0%)	0.796
0	18(34.6%)	21(42.0%)	
1	11(21.2%)	8(16.0%)	
2	2(3.8%)	1(2.0%)	
3			
ROP exam time	81.92(15.72)	78.58(17.74)	0.316
Mean(SD)	40–110	45–120	
Min – Max			

Postprocedure apnea occurred in 17.3% of the infants in the microdrop group and 24.0% in the standard group ($P = 0.40$). No cases of necrotizing enterocolitis or serious adverse ocular events were reported.

All examinations were successfully completed, with a 100% procedural success rate in both groups. The distributions of ROP stages and examination durations were similar between the groups, with mean (SD) examination times of 81.9 (15.7) minutes and 78.6 (17.7) minutes, respectively ($P = 0.32$).

There was no difference in the secondary outcomes between the two subgroups, as shown in additional Tables 5, 6, 7, 8, 9, 10, 11 and 12.

Discussion

This randomized controlled noninferiority trial demonstrated that compared with standard drop instillation, microdrop administration of 2.5% phenylephrine and 0.8% tropicamide results in noninferior pupillary dilation. All examinations were successfully completed with comparable mydriatic efficacy at 45, 90, and 120 minutes post-instillation. No significant differences in heart rate, oxygen saturation, or blood pressure were observed between the groups. This finding extends emerging evidence supporting reduced-volume mydriatic regimens as safe, effective alternatives to standard dosing in preterm infants[7, 8].

Three recent randomized controlled trials now provide convergent evidence for microdrop efficacy. The Little Eye Drop Study demonstrated that very low-dose microdrops were noninferior to low-dose regimens in preterm infants[7]. Compared with standard drop instillation, the MyMiROPS trial showed superior mydriatic efficacy at 45 minutes and reduced oxygen desaturation following microdrop instillation[18]. Our findings using 2.5% phenylephrine demonstrate that the principle of volume reduction applies across a wide range of drug concentrations. The 2019 systematic review by Kremer et al. revealed that published evidence supporting current mydriatic guidelines was largely underpowered with a high risk of bias, despite the widespread use of adult-equivalent dosing in neonatal units[1]. The convergence of recent trials across geographies and formulations validates that reduced-volume delivery represents a fundamental principle for optimizing the risk–benefit profile of mydriatic therapy. The safety advantage of microdrop administration is grounded in neonatal pharmacology. Standard drops (approximately 26 μ L) substantially exceed neonatal tear film capacity (0.5–2 μ L), resulting in excessive nasolacrimal drainage and systemic absorption[3]. Up to 80% of instilled mydriatics may undergo systemic absorption, which is particularly significant in neonates because of their low circulating blood volume and immature hepatic metabolism[1, 2]. Microdrops (5–15 μ L) more closely approximate physiological tear film volumes, optimizing the local drug effect while minimizing systemic exposure[4–6]. Both groups achieved mean pupillary diameters exceeding 6 mm at 45 minutes, which is well above the threshold required for adequate retinal visualization. All examinations were successfully completed, including identification of treatment-requiring ROP. These findings confirm that a reduction in drop volume does not compromise diagnostic efficacy[7–8].

Physiological monitoring revealed no significant differences in adverse effects between the groups. Postprocedure apnea occurred in 17.3% of the patients in the microdrop group and 24.0% of those in the standard group. No gastrointestinal complications or serious adverse events were observed. These findings are consistent with previous studies documenting the physiological instability associated with ROP examinations[19]. Unlike earlier microdrop studies requiring micropipette delivery, the use of a bottle adapter system demonstrated feasibility for routine clinical practice. This simplifies administration

and enhances applicability across diverse neonatal settings, particularly in resource-limited environments.

This investigation employed rigorous methodology, including randomized allocation with concealment, blinded outcome assessment, stratification by gestational age, and a prospectively calculated sample size, which is consistent with established standards for noninferiority trials[15, 17]. However, this single-center study may not fully represent other healthcare settings. Although adequately powered for the primary outcome, the sample size may be insufficient to detect rare adverse events. Manual measurement of pupillary diameter introduces potential error compared with automated methods; the use of photograph-based pupillometry in prior studies represents a methodological improvement[7]. The lack of pharmacokinetic analysis limits mechanistic interpretation; however, prior studies have demonstrated increased systemic bioavailability with standard drops, supporting volume reduction strategies[18].

Current evidence suggests that transitioning from standard large-volume drops to reduced-volume microdrops is a preferred approach for neonatal ROP screening[7, 8, 18]. This aligns with clinical guidelines emphasizing safe and effective mydriatic use in preterm infants[20]. Future multicenter trials with larger sample sizes are needed to detect rare adverse events and assess cumulative exposure across repeated ROP examinations. Comparative effectiveness studies evaluating clinically meaningful outcomes, including necrotizing enterocolitis, sepsis, and mortality, would provide stronger evidence for widespread adoption.

CONCLUSION

Microdrop administration of phenylephrine and tropicamide via a bottle adaptor achieved mydriasis comparable with that of standard drops, without evidence of increased systemic adverse effects. These findings support the use of reduced-volume mydriatic regimens as **safe, effective, and scalable alternatives** for ROP screening in preterm infants.

Abbreviations

- **CI**

Confidence interval

- **DBP**

Diastolic blood pressure

- **GA**

Gestational age

- **HR**

Heart rate

- **MBP**

Mean blood pressure (mean arterial blood pressure)

- **NEC**

Necrotizing enterocolitis

- **NICU**

Neonatal intensive care unit

- **ROP**

Retinopathy of prematurity

- **SBP**

systolic blood pressure

- **SD**

Standard deviation

- **SpO₂**

Oxygen saturation

- **T45**

45 minutes after the first drop instillation

- **T90**

90 minutes after the first drop instillation

- **T120**

120 minutes after the first drop instillation

Declarations

Ethics approval and consent to participate

The study protocol was approved by the Institutional Ethics Committee of Madras Medical College, Chennai (IEC No. MMC/Approval/12052025). The trial was registered with the Clinical Trials Registry of India (CTRI/REF/2025/03/101681) on 10-03-2025. Written informed consent was obtained from the parents or legal guardians prior to enrollment. The study was conducted in accordance with the Declaration of Helsinki.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Authors' information

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Author Contribution

VE (Vivetha Elango): Conceived and designed the study, coordinated and conducted data collection, performed data interpretation, drafted the manuscript, and is responsible for overall study integrity and accuracy of the work. NE (Narayanan E): Contributed to study design, assisted with data collection and interpretation, and critically revised the manuscript for important intellectual content. MN (Muthukumaran N): Supervised study conduct, provided methodological and clinical oversight, contributed to data interpretation, and critically reviewed and approved the final manuscript. All authors read and approved the final version of the manuscript and agree to be accountable for all aspects of the work.

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Not applicable.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author at https://docs.google.com/spreadsheets/d/1cJv6CEACH96jNTUsKQIZFKEo9Ose_leN/edit?usp=sharing&ouid=100838261379668675094&rtpof=true&sd=true

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Figures



Figure 1

Bottle adapter (Nanodropper) used to deliver microdrops

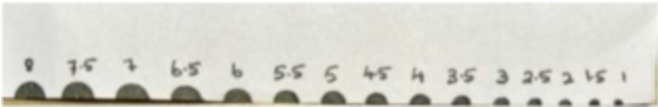


Figure 2

Customized ruler used to measure pupillary diameter

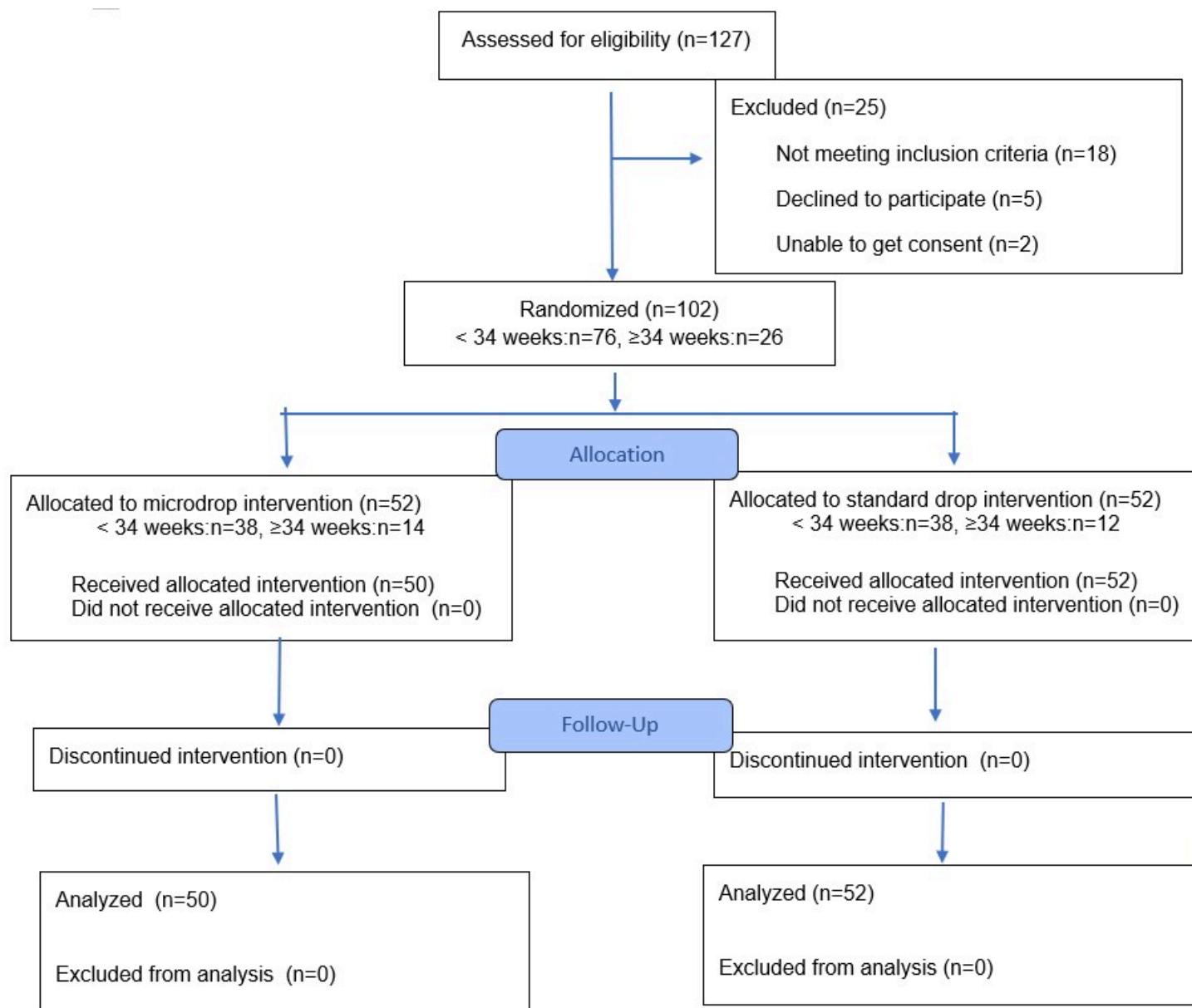


Figure 3

CONSORT Participant flow diagram

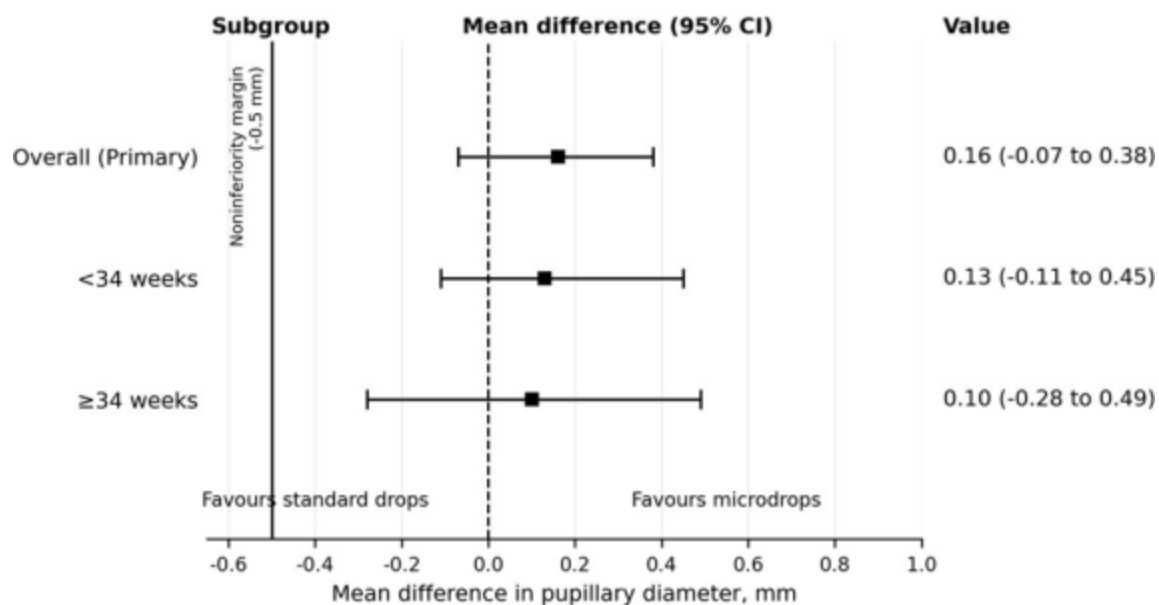


Figure 4

Forest plot of the primary outcome and gestational age subgroup analysis

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