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Critical BCVA Threshold and Refractive Error Modulation for Fixation Instability in Strabismic Amblyopia: Revealed by Eye-Tracking and GAM Modeling

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Abstract

Purpose

To determine the critical best-corrected visual acuity (BCVA) threshold associated with binocular fixation instability in children with strabismic amblyopia, and to evaluate the modulatory effect of refractive error on this sensory–motor coupling using high-resolution eye tracking and nonlinear modeling.

Methods

In this multicenter case-control study, 536 children with amblyopia and 159 sex-matched controls were enrolled from the China Amblyopia Behavior Cohort (CABC, 2024–2025). Binocular fixation stability (primary outcome: targeting displacement [TD]) was measured with a 133 Hz Tobii Eye Tracker 4. LOESS smoothing and generalized additive models (GAMs) were applied to characterize nonlinear BCVA–TD relationships. Tensor product interactions tested the moderating effect of spherical equivalent (SE).

Results

All amblyopia subtypes exhibited significantly greater fixation instability compared with controls (all $P < 0.001$). A nonlinear BCVA–TD relationship was observed exclusively in strabismic amblyopia, with fixation instability increasing sharply beyond a BCVA threshold of 0.321 logMAR (effective degrees of freedom, 4.46; $P = 0.002$). Higher SE amplified BCVA-related TD increases in ametropic ($P = 0.013$) and strabismic ($P = 0.004$) subtypes, demonstrating refractive error modulation of sensory-motor coupling. The interaction model explained 27.9% of TD variance (adjusted $R^2 = 0.251$).

Conclusions

Strabismic amblyopia shows abrupt fixation instability beyond 0.321 logMAR; refractive error exacerbates this in ametropic/strabismic subtypes. Fixation stability should be a routine biomarker, with optimized refractive correction before strabismic BCVA falls below this threshold.

Introduction

Amblyopia, a neurodevelopmental disorder affecting 2%–4% of children worldwide, is characterized by reduced best-corrected visual acuity (BCVA) in the absence of structural ocular abnormalities and despite optimal optical correction [1–3]. Beyond impairing spatial vision and stereopsis, amblyopia is increasingly recognized as a disorder of oculomotor control. Binocular fixation instability—quantified as targeting displacement (TD) during sustained gaze—has emerged as a key biomarker of sensory–motor dysfunction [4–6]. Importantly, fixation instability manifests in a subtype-specific manner: strabismic

amblyopia is associated with excessive fixation drift due to ocular misalignment, while anisometropic amblyopia shows a progressive increase in TD with declining acuity [7, 8]. Such instability exacerbates visual blur, impedes information acquisition, and correlates strongly with poor stereopsis and reading performance [5, 9], underscoring its clinical relevance for prognosis and rehabilitation [10].

Despite these insights, several knowledge gaps remain. First, most prior studies have relied on monocular recordings [6, 9, 11], thereby neglecting natural binocular viewing conditions in which interocular interactions strongly influence fixation control [12]. Second, the relationship between BCVA and fixation instability has often been modeled as linear [13, 14], overlooking potential threshold effects suggested in strabismic amblyopia [4]. Third, the role of refractive error, particularly spherical equivalent (SE), as a moderator of sensory–motor coupling remains poorly understood. Evidence indicates that uncorrected refractive error alters microsaccade dynamics and spatial processing [15, 16], but whether SE interacts with BCVA to drive fixation collapse in different amblyopia subtypes is unknown.

To address these gaps, we leveraged data from the China Amblyopia Behavior Cohort (CABC) to investigate binocular fixation stability using high-resolution eye-tracking. We applied generalized additive models (GAMs) to: (1) identify a critical BCVA threshold associated with fixation instability in strabismic amblyopia; and (2) test whether refractive error modulates sensory-motor coupling (BCVA–TD association) in a subtype-specific manner.

We hypothesized that: BCVA degradation beyond a functional threshold would disproportionately disrupt fixation stability in strabismic amblyopia due to compromised cortical integration of misaligned inputs [7, 17]; Higher SE would amplify BCVA-driven TD increases in ametropic and strabismic subtypes by exacerbating optical defocus and neural noise [1, 16]. Therefore, this study aims to delineate the precise, potentially non-linear, relationship between BCVA and fixation instability under binocular viewing conditions, and to elucidate how refractive error modulates this sensory-motor coupling across different amblyopia subtypes. These findings aim to inform subtype-stratified rehabilitation targeting both sensory and motor deficits.

Methods

1. Study Design and Participants

This multicenter case-control study analyzed baseline data from the China Amblyopia Behavior Cohort (CABC, 2024–2025). A total of 536 children with clinically confirmed amblyopia (303 ametropic, 201 anisometropic, 32 strabismic) and 159 sex-matched controls were recruited from three tertiary eye hospitals in Guangxi, Guangdong, and Sichuan, China.

Amblyopia subtypes were defined according to the Chinese Ophthalmological Society guidelines [18]: (1) Ametropic amblyopia: Bilateral BCVA reduction with interocular SE difference < 0.75 D; (2) Anisometropic

amblyopia: Interocular SE difference ≥ 1.50 D or cylindrical difference ≥ 1.00 D; (3) Strabismic amblyopia: amblyopia associated with manifest strabismus, confirmed by prism cover test (> 8Δ) [19].

Control participants had normal BCVA (logMAR ≤ 0.10 in both eyes), no history of amblyopia, ocular surgery, or neurological disease. Sample size was determined using a priori power analysis (target power 0.90, α = 0.05, effect size f^2 = 0.05), Specifically, the effect size f^2 = 0.05 was determined based on relevant literature, indicating a minimum of 335 participants to detect nonlinear effects in GAMs [20].

The study adhered to the Declaration of Helsinki and was approved by the Ethics Committee of the People's Hospital of Guangxi Zhuang Autonomous Region (KY-KJT-2023-285). Written informed consent was obtained from all legal guardians.

2. Ophthalmic Examination Protocol

All participants underwent standardized ophthalmic examinations and stereoacuity assessment (Fig. 1). BCVA was measured using a standard logarithmic visual acuity chart at 5 m, the result was recorded via the LogMAR scale. Anterior segment and fundus evaluation were examined using slit-lamp biomicroscopy and ophthalmoscopy. Ocular alignment was assessed using prism cover test (strabismus: median 19.69Δ, range from 8.75 ~ 43.75Δ) and fixation behavior assessment. Cycloplegic refraction was performed 30–40 min after instillation of 1% atropine sulfate (three drops at 10-min intervals). SE was calculated as sphere + 0.5 × cylinder.

Fine stereoacuity was measured with the Brain Imaging Bio-Stimulation Training System (National Engineering Research Center for Medical Devices, China). Participants, seated 80 cm from the display and wearing polarized glasses, identified the orientation of E-shaped targets (3° × 3°). Fine stereoacuity thresholds were graded logarithmically: grade 0 (2.90), grade 1 (2.60), grade 2 (2.48), grade 3 (2.30), grade 4 (2.00), with higher grades indicating better fine stereoacuity.

3. Binocular Fixation Stability Assessment

3.1 Eye-Tracking System

Binocular fixation stability was measured using the Tobii Eye Tracker 4 (Tobii 4C, Sweden), sampling at 133 Hz with average spatial accuracy of 0.5–1.0° and precision of 0.03° RMS. Participants were seated 80 cm from a 23-inch monitor (1920×1080 pixels) in a dimly lit, sound-attenuated room. A nine-point binocular calibration was performed prior to each task, with calibration error required to be < 0.5°.

3.2 Sustained Fixation Task

A high-contrast yellow circular target (10° diameter) was presented sequentially at eight peripheral locations (± 5° horizontally and vertically) and centrally. Each stimulus lasted 2 s, with 0.5 s transition

intervals. Participants were instructed to maintain fixation while minimizing head movement. Gaze coordinates were recorded in pixels.

3.3 Targeting Displacement (TD)

Binocular fixation stability was quantified as TD. TD is calculated as the average offset from the fixation point to the target center per frame, yielding horizontal (Fix_X), vertical (Fix_Y), and average TD values for each trial (Fig. 1). Targeting Displacement (TD) Calculated using the formula:

$$\text{TD}= \frac{1}{\text{N}}\sum_{i=1}^{N}\sqrt{(x_i - x_c)^2 + (y_i - y_c)^2}$$

Where (x_i, y_i) represents the binocular gaze coordinates for sample i, and (x_c, y_c) denotes the coordinates of the target center. This metric is a standard measure of fixation dispersion [21, 22]. Data segments with an eye tracking loss rate > 20% are excluded from analysis to ensure data quality [23].TD represents spatial dispersion of binocular gaze points, conceptually similar to the dispersion component of the bivariate contour ellipse area (BCEA). Unlike BCEA, which models a 68–95% probability ellipse based on variance and covariance, TD reflects frame-wise Euclidean deviation and has been widely used in pediatric and low-vision eye-tracking studies. As BCEA requires longer stable fixation intervals, TD was selected for the current design involving brief multi-position fixations.

4. Statistical Modeling of Sensory-Motor Coupling

LOESS smoothing (span = 0.75) was used for exploratory visualization of BCVA–TD relationships by subtype. Nonlinear effects were tested using GAMs with thin-plate regression splines:

TD ~ s (BCVA, by = subtype) + age + sex + SE + stereoacuity

To assess moderation by refractive error, tensor product interactions were modeled:

TD ~ ti (BCVA, SE, by = subtype) + covariates

Significance of smooth and interaction terms was determined using false discovery rate (FDR)–corrected P values. Threshold identification was using first-derivative analysis of marginal effects. Robustness was validated through bootstrap resampling (n = 1,000) in the strabismic subgroup, 95% CI for effective degrees of freedom (edf), proportion of P < 0.01 resamples were reported. Analyses were conducted in R (version 4.5.1; mgcv package), β coefficients and 95% CIs reported for all models.

Results

1. Clinical Characteristics

A total of 695 participants were included: 536 children with amblyopia (303 ametropic, 201 anisometropic, 32 strabismic) and 159 controls. Demographic and clinical data are summarized in Table 1. Significant group differences were observed for age, SE, BCVA, stereoacuity, and fixation stability (all P < 0.001). Children with strabismic amblyopia exhibited the poorest BCVA (median logMAR 0.60) and highest SE (median + 4.8 D).

Table 1
Demographic and Clinical Characteristics of Amblyopia Subtypes and Controls

Variable	Total (N = 695).	Ametropic (n = 303)	Anisometropic (n = 201)	Strabismic (n = 32)	Control (159)	P value
Gender, n (%)						0.760
Boys	339 (48.8)	154 (50.8)	97 (48.3)	14 (43.8)	74 (46.5)	
Girls	356 (51.2)	149 (49.2)	104 (51.7)	18 (56.2)	85 (53.5)	
Age, Median (IQR)	6.0 (5.0, 7.0)	6.0 (5.0, 6.0)	6.0 (5.0, 6.0)	6.0 (5.0, 6.0)	7.0 (6.0, 9.0)	< 0.001
SE, Median (IQR)	0.6 (0.0, 3.5)	0.9 (-0.2, 3.5)	2.5 (0.4, 4.5)	4.8 (0.9, 6.7)	0.0 (0.0, 0.0)	< 0.001
BCVA, Median (IQR)	0.3 (0.2, 0.4)	0.3 (0.2, 0.4)	0.4 (0.2, 0.5)	0.4 (0.3, 0.5)	0.0 (0.0, 0.1)	< 0.001
Fine stereopsis Median (IQR)	2.3 (2.0, 2.9)	2.3 (2.0, 2.9)	2.9 (2.3, 2.9)	2.9 (2.6, 2.9)	2.0 (2.0, 2.0)	< 0.001
Fix_X Median (IQR)	116.7 (61.8, 216.7)	133.4 (70.7, 233.8)	115.2 (69.9, 197.7)	301.7 (142.0, 643.6)	76.3 (45.4, 154.3)	< 0.001
Fix_Y, Median (IQR)	116.4 (75.7, 176.8)	130.0 (81.7, 189.3)	123.0 (82.4, 175.6)	160.0 (111.4, 208.8)	83.3 (59.3, 131.6)	< 0.001
Fixation_TD, Median (IQR)	121.4 (72.2, 198.4)	141.1 (81.8, 212.0)	124.9 (81.8, 189.3)	257.9 (143.8, 487.7)	84.9 (58.0, 144.2)	< 0.001

Values are presented as median (interquartile range, IQR) or number (percentage). SE = spherical equivalent; BCVA = best-corrected visual acuity; Fix_X = horizontal targeting displacement; Fix_Y = vertical targeting displacement; TD = targeting displacement. P values are from Kruskal–Wallis or χ^2 tests as appropriate.

2. Subtype-Specific Fixation Instability

Binocular fixation instability, quantified as TD, was significantly greater in all amblyopia subtypes compared with controls (all $P < 0.001$). The magnitude of instability differed by subtype, median TD difference was + 40.3px [95% CI: 25.7–55.7] in ametropic amblyopia, + 32.1px [19.6–45.6] in anisometropic amblyopia, and + 149.3px [92.7–213.5] in strabismic amblyopia respectively (all $P < 0.001$; Fig. 2). The largest effect was observed in strabismic amblyopia, which showed marked deficits in horizontal (Fix_X), vertical (Fix_Y), and mean TD (all $P < 0.001$ vs. other subtypes).

3. Nonlinear BCVA–TD Association in Strabismic Amblyopia

LOESS plots revealed a distinct threshold-dependent relationship exclusively in strabismic amblyopia (Fig. 3). While ametropic, anisometropic, and control groups showed flat BCVA–TD curves, strabismic cases exhibited accelerated TD escalation when BCVA exceeded 0.3 logMAR.

4. GAMs Ananlysis and Threshold Identification

GAM analysis confirmed a significant nonlinear association between BCVA and TD in strabismic amblyopia (edf = 4.46, $P = 0.002$). Marginal effect analysis identified a critical inflection point at 0.321 logMAR (Fig. 4A), beyond which TD increase sharply. Specifically, each 0.1 logMAR decrease in acuity was associated with an additional 38.2 px increase in TD (95% CI: 25.7–50.7; Fig. 4).

5. Refractive Error Modulation

Tensor product interactions indicated that higher SE amplification BCVA-related increases in fixation instability (TD) in ametropic amblyopia (β = +17.3 px/D per logMAR unit, $P = 0.013$) and strabismic (β = +42.6 px/D per logMAR unit, $P = 0.004$). The interaction model explained 27.9% of TD variance (adjusted $R^2 = 0.251$), outperforming the main-effect model (Δdeviance explained = 4.6%, $P < 0.001$; Table 2).

Table 2
Comparison of Parametric and Smooth Effects Between the Main Effect and Interaction Models

Variable	**Model1**		**Model2**	
Parametric Terms	Estimate	p-value	Estimate	p-value
(Intercept)	103.490	0.003	110.071	0.001
Age	-11.250	< 0.001	-10.551	< 0.001
Gender				
Boys	0(ref)		0(ref)	
Girls	-23.728	0.004	-21.803	0.008
SE	2.341	0.155	—	—
Fine stereopsis	57.987	< 0.001	52.618	< 0.001
Type_group				
Ametropic	0(ref)		0(ref)	
Anisometropic	-28.015	0.057	-23.129	0.111
Strabismic	199.350	< 0.001	215.116	< 0.001
Control	-22.227	0.268	-19.070	0.334
Smooth Terms	edf	p-value	edf	p-value
s(BCVA): Ametropic	3.764	0.404	3.732	0.228
s(BCVA): Anisometropic	1.001	0.472	1.001	0.610
s(BCVA): Strabismic	4.460	0.002	4.473	0.002
s(BCVA): Control	1.002	0.472	1.001	0.622
s(SE)	—	—	1.585	0.524
Interaction Terms	edf	p-value	edf	p-value
ti(BCVA, SE): Ametropic	—	—	2.708	0.013
ti(BCVA, SE): Anisometropic	—	—	1.001	0.977
ti(BCVA, SE): Strabismic	—	—	4.703	0.004
ti(BCVA, SE): Control	—	—	0.001	0.977

Parametric terms represent fixed effects. Smooth terms are modeled using thin-plate regression splines. Interaction terms represent tensor product interactions between BCVA (best-corrected visual acuity) and SE (spherical equivalent). Model 1 = main-effect model; Model 2 = interaction model. Adjusted R^2 and deviance explained reflect overall model performance. Significant effects are bolded.

Variable	Model1	Model2
R-sq.(adj)	0.213	0.251
Deviance explained	23.3%	27.9%

Parametric terms represent fixed effects. Smooth terms are modeled using thin-plate regression splines. Interaction terms represent tensor product interactions between BCVA (best-corrected visual acuity) and SE (spherical equivalent). Model 1 = main-effect model; Model 2 = interaction model. Adjusted R^2 and deviance explained reflect overall model performance. Significant effects are bolded.

6. Bootstrap Validation of Nonlinear Effects

In the strabismic subgroup (n = 32), bootstrap resampling (n = 1,000 iterations) confirmed robustness of nonlinear effect: the 95% CI for edf of the BCVA smooth term ranged from 3.1–7.2, and 38.7% of resamples showed $P < 0.01$ for nonlinearity.

7. Sensory-Motor Decoupling Beyond the Threshold

Marginal effect curves illustrated an abrupt shift in fixation stability in strabismic amblyopia at the identified BCVA threshold (Fig. 5). Below 0.321 logMAR, TD remained stable (slope = -5.2 px/logMAR). Above the threshold, TD increased steeply (slope = + 49.1 px/logMAR, 95% CI: +32.7–+65.5), consistent with a collapse of binocular fixation control.

Discussions

1. Principal Findings

This study identified a critical BCVA threshold of 0.321 logMAR (≈ 20/42 Snellen), beyond which binocular fixation stability deteriorated sharply in children with strabismic amblyopia. Refractive error further amplified this sensory–motor decoupling in both ametropic and strabismic subtypes. These findings extend previous work by demonstrating that fixation instability is not merely proportional to acuity loss, but instead follows a threshold-dependent trajectory in strabismus.

2. Integration with Previous Research

Our results confirm and refine prior observations that fixation instability is a hallmark of amblyopia [6, 7]. Subramanian et al. [6] reported reduced fixation stability in amblyopic children using monocular recordings, while Kelly et al.[7] demonstrated greater instability in binocular conditions. By applying nonlinear modeling, we show that strabismic amblyopia is uniquely characterized by an abrupt inflection point, consistent with Birch's hypothesis of cortical integration failure when acuity loss surpasses a

functional limit [4]. This may explain why some smaller studies failed to detect clear associations between acuity and fixation [14]—linear models likely masked threshold effects.

We also found that higher SE exacerbates fixation instability in ametropic and strabismic amblyopia, consistent with Wahl et al.'s proposal that optical defocus increases neural noise and disrupts microsaccade control [16]. In contrast, anisometropic amblyopia showed little BCVA–TD coupling, unlike strabismic amblyopia caused by ocular misalignment and abnormal dorsal oculomotor circuits, anisometropic amblyopia is mainly characterized by interocular aniseikonia and asymmetric retinal image quality, rather than strabismus [24]. Long-term binocular perceptual imbalance during visual development leads to specific cortical remodeling and compensatory fixation control in anisometropic amblyopia [12]. instead of increased neural noise in oculomotor pathways. Combined with preserved ocular alignment [8], this subtype-specific mechanism may explain the weak BCVA–TD coupling in anisometropic amblyopia. Taken together, these findings indicate that fixation instability in amblyopia is not uniform, but varies by subtype, acuity threshold, and refractive factors; this threshold-dependent pattern likely reflects underlying cortical and oculomotor mechanisms.

3. Neurobiological Interpretation

Building on the threshold-dependent pattern observed in strabismic amblyopia, the identified threshold at 0.321 logMAR likely reflects a transition in cortical control of fixation. Below this level, residual binocular input may stabilize gaze through dorsal stream regulation of the superior colliculus [25]. Beyond the threshold, imbalanced input to V1/V2 disrupts efferent copy signals to frontal eye fields, precipitating fixation collapse [26, 27]. Functional MRI studies in amblyopia have documented abnormal parvocellular pathway activation and impaired integration across extrastriate areas [28, 29], supporting this interpretation.

4. Clinical Translation and Rehabilitation Implications

These findings have several clinical applications. (1) Threshold-Guided Intervention: For strabismic amblyopia with BCVA worse than 0.321 logMAR, early refractive correction is critical to mitigate SE-related amplification of instability (β = +42.6 px/D), followed by binocular integration training (e.g., dichoptic motion tasks) to restore sensory–motor integration [27]. (2) Subtype-Specific strategies: Strabismic amblyopia may benefit from gaze stabilization therapy (Counteracts threshold-triggered collapse), ametropic amblyopia from high-frequency spatial training to reduce neural noise [16], and anisometropic amblyopia from contrast-balanced binocular approaches [8, 30]. (3) Prognostic value: Fixation stability could serve as a biomarker of disease severity and treatment responsiveness, aligning with recent efforts to incorporate oculomotor endpoints in amblyopia trials [27, 31].

5. Methodological Advances

This study introduces several methodological strengths. First, binocular eye-tracking captures naturalistic fixation control, addressing limitations of earlier monocular paradigms [9]. Second, GAMs reveal nonlinear threshold effects that conventional regression approaches cannot detect [32]. Third, interaction modeling demonstrated for the first time that refractive error modulates fixation instability in a subtype-dependent manner ($P_{interaction} < 0.01$). Together, these advances support the utility of nonlinear modeling in pediatric vision science.

6. Limitations and Future Directions

Several limitations warrant consideration. The cross-sectional design precludes causal inference regarding whether acuity decline precedes fixation collapse. Longitudinal data from the ongoing CABC study will be essential to address temporal dynamics. In addition, the relatively small strabismic sample (n = 32) requires replication in larger cohorts, although bootstrap validation supports robustness. Finally, the current study only utilized a spatial dispersion metric TD (to quantify binocular fixation instability), which provides a limited view of oculomotor function. To generate a more comprehensive oculomotor profile and refine mechanistic interpretations, future work should incorporate additional oculomotor metrics such as microsaccade frequency and drift velocity to refine mechanistic interpretation.

Declarations

Conflicts of Interest

The author(s) declare that there are no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Author Contribution Statement

Conceptualization: J.L., J.Z., X.X.; Methodology:J.L., J.Z., X.X.; Formal analysis: M.R.; Investigation: L.P., Y.H., W.L., L.L., Y.L., E.L., M.K., Q.C., Y.L.; Data curation: L.P., M.R., Y.H., L.L., Y.L., Q.C., Y.L.,E.L.; Validation: W.L., M.K., Q.C.; Writing-original draft: All authors; Writing-review & editing:J.L., J.Z., X.X.; Funding acquisition:J.L., J.Z., X.X

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Figures

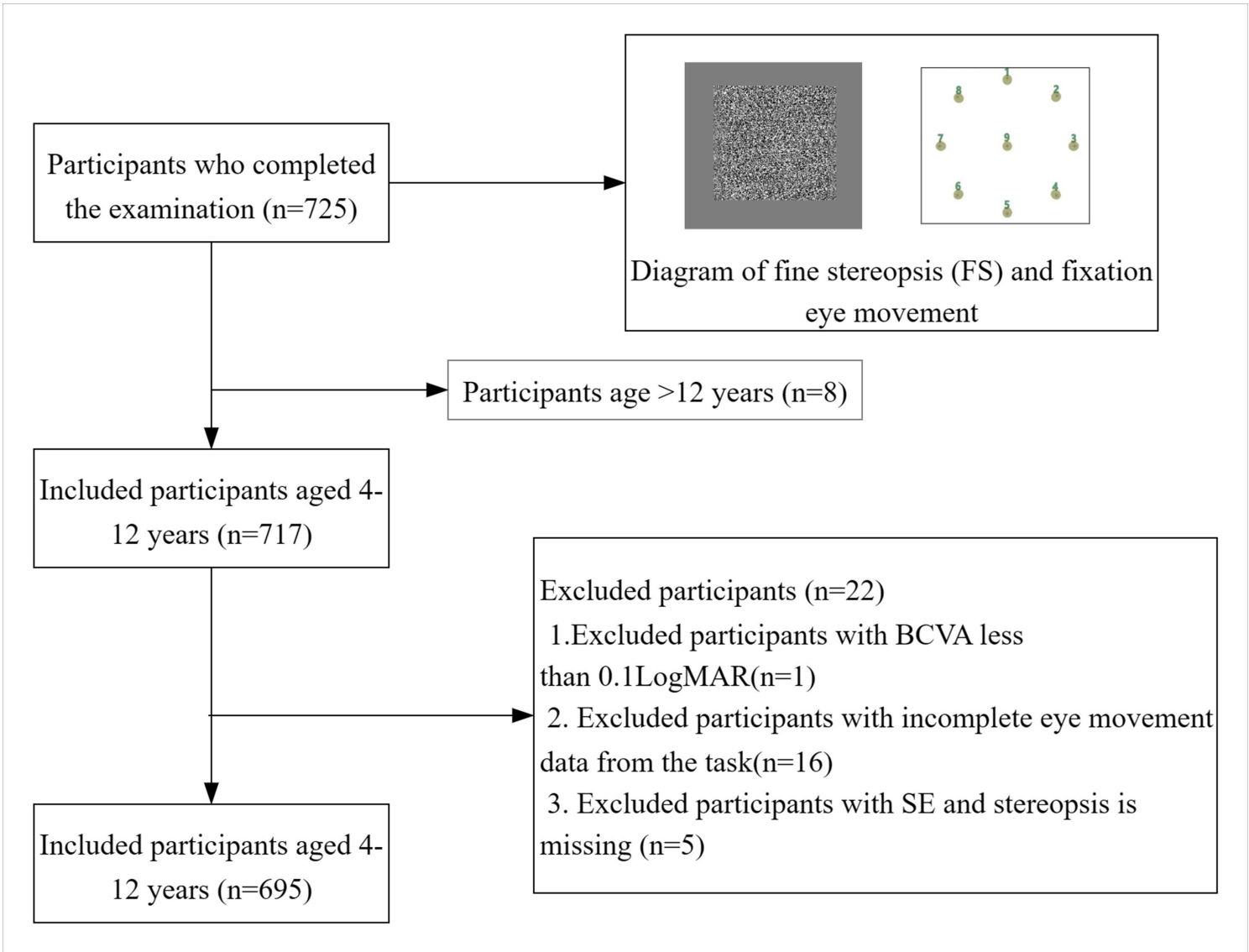


Figure 1

Flowchart of participant enrollment in the China Amblyopia Behavior Cohort (CABC).

This flow diagram illustrates the recruitment and selection of children with ametropic, anisometropic, and strabismic amblyopia, as well as control participants, included in the final analysis.

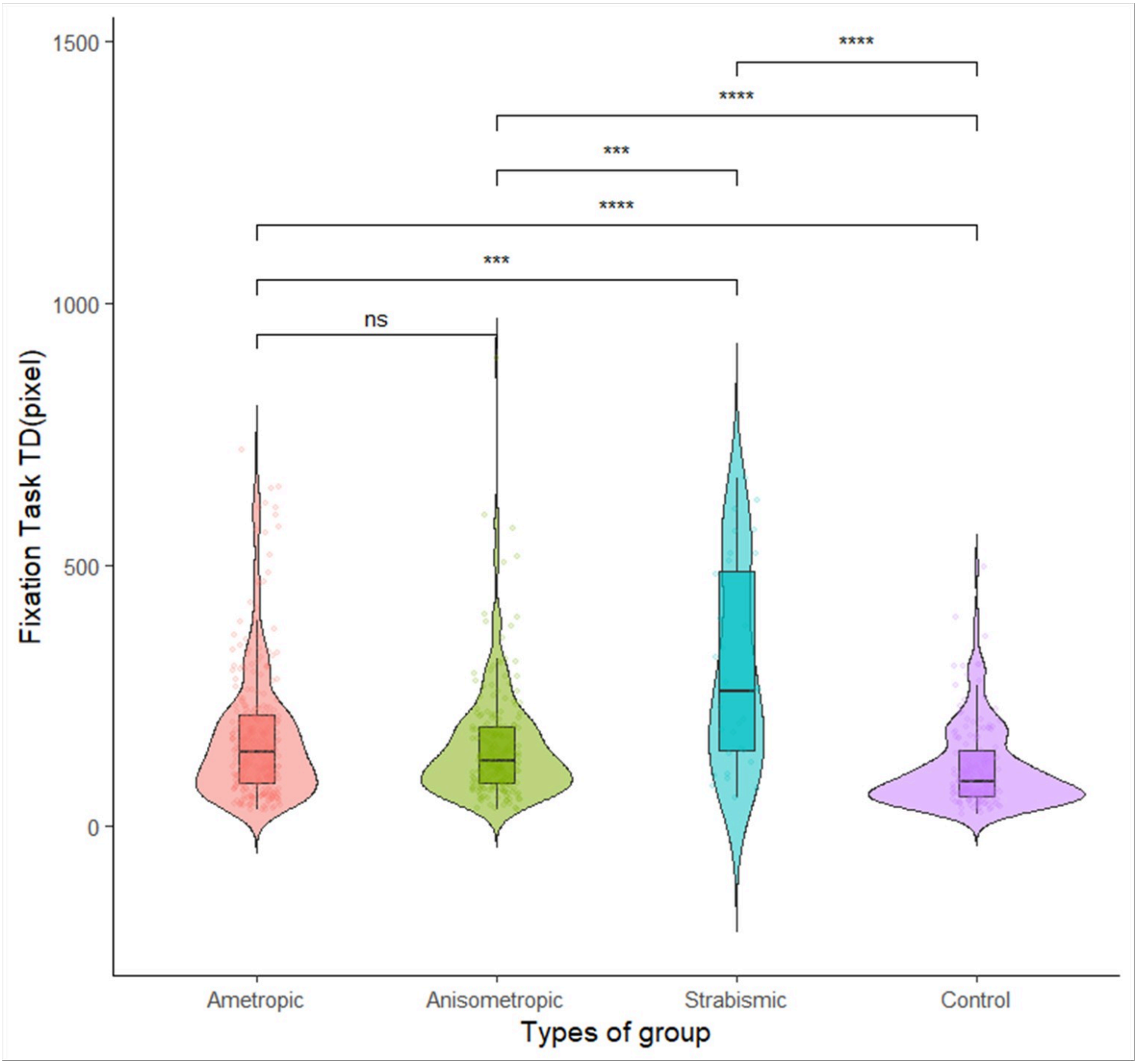


Figure 2

Distribution of fixation instability across study groups.

Box plots show targeting displacement (TD) during sustained fixation tasks across amblyopia subtypes and controls. TD = average offset between gaze position and target center. Higher TD indicates greater fixation instability.

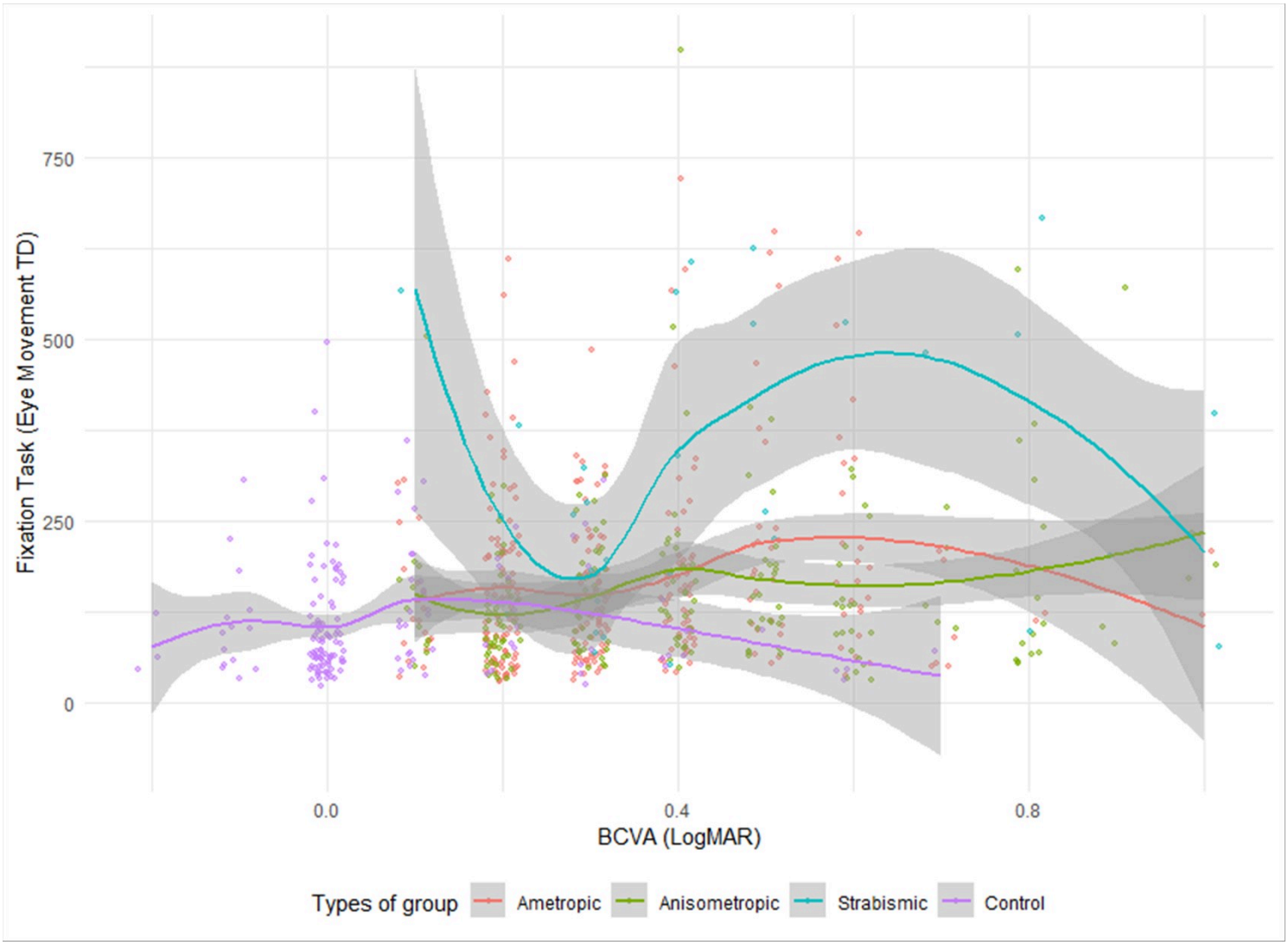


Figure 3

Nonlinear associations between BCVA and fixation instability.

LOESS smoothing plots of TD (targeting displacement) against BCVA (best-corrected visual acuity) across groups. A threshold-like nonlinear increase in TD is observed only in the strabismic amblyopia group.

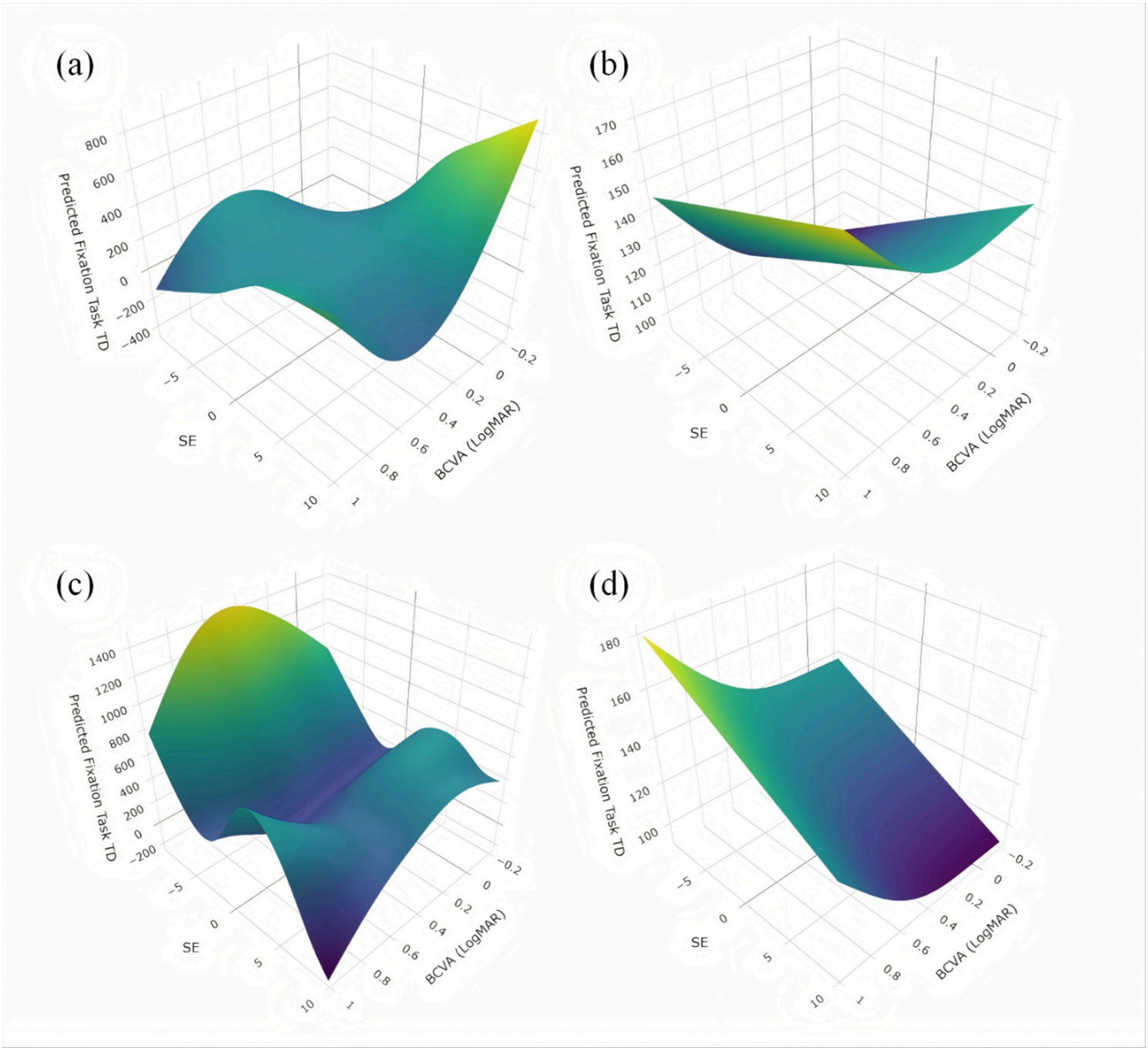


Figure 4

Interaction between BCVA and refractive error in predicting fixation instability.

Generalized additive model (GAM) interaction surfaces depicting the moderating effect of spherical equivalent (SE) on the relationship between BCVA and TD (targeting displacement) in (a) ametropic amblyopia, (b) anisometropic amblyopia, (c) strabismic amblyopia, and (d) controls. Warmer colors indicate higher TD values.

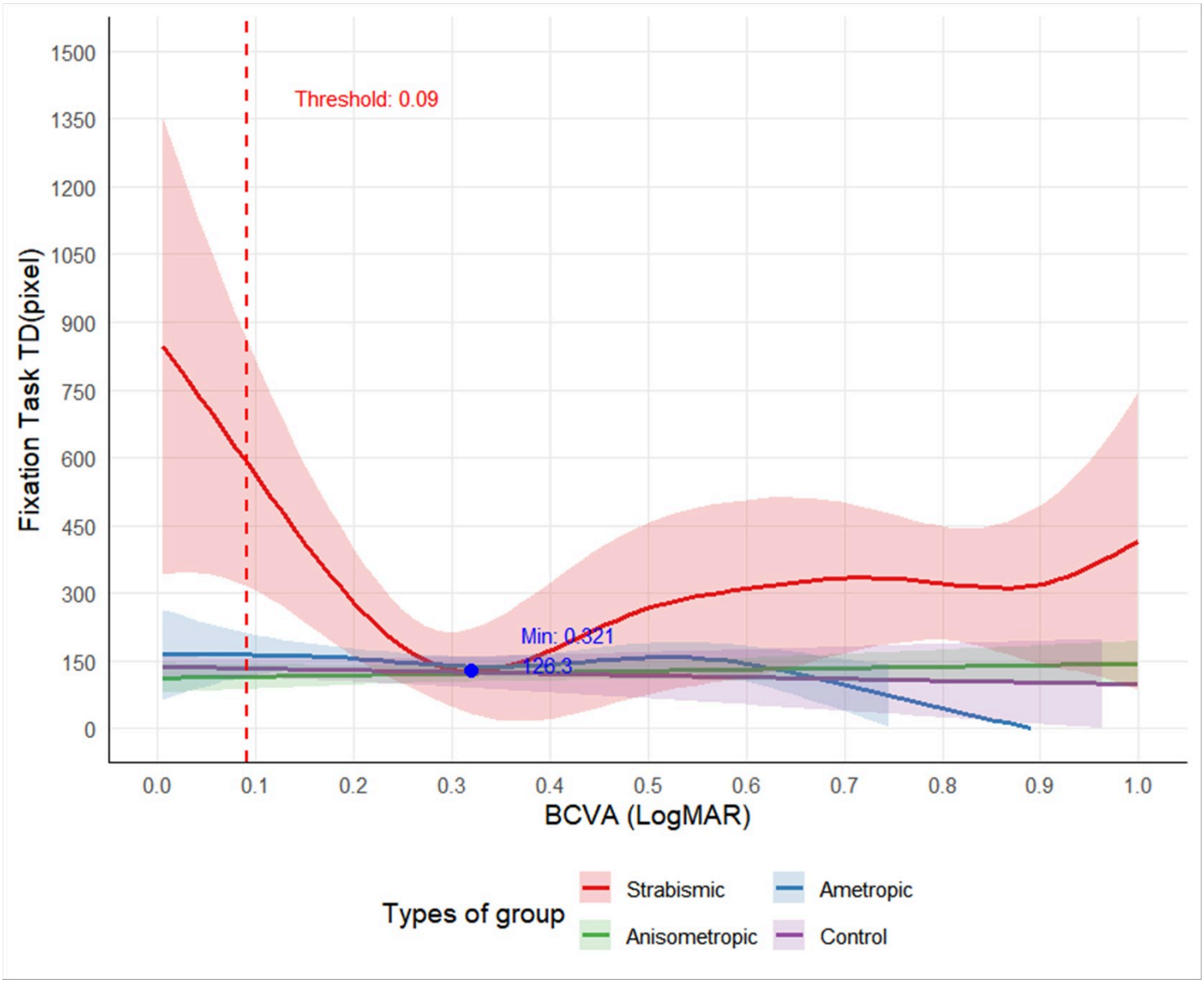


Figure 5

Predicted fixation instability across levels of visual acuity in strabismic amblyopia.

Marginal effect curves from GAM analysis. Below the threshold of 0.321 logMAR, fixation stability remains relatively preserved. Beyond this threshold, targeting displacement (TD) escalates sharply, indicating collapse of binocular fixation control.