

# Assessment of Retinal Nerve Fiber Layer Thickness Strabismic and Anisometropic Amblyopia Cases

## Şaşılık ve Anizometropik Ambliyopi Olgularında Retinal Sinir Lifi Tabaka Kalınlığının Değerlendirilmesi

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### Abstract

**Objective:** The aim of this study was to determine whether differences exist in retinal nerve fiber layer (RNFL) thickness between amblyopic eyes and non-amblyopic eyes of patients with strabismic amblyopia and anisometropic amblyopia using optical coherence tomography (OCT), as well as healthy eyes in a control group.

**Method:** Thirty-six cases of unilateral anisometropic amblyopia, 40 cases of unilateral amblyopia with strabismus, and 42 healthy individuals were included. Patient history, demographic characteristics, and mean RNFL thickness values across quadrants were recorded using OCT.

**Results:** A total of 118 participants were enrolled in the study, comprising 54 males and 64 females, with an average age of 16.25±9.05 years. Analysis of mean and quadrant RNFL thickness values revealed no statistically significant differences between amblyopic eyes, non-amblyopic eyes, and the control group ( $p>0.05$ ). Similarly, there was no significant variation in RNFL thickness in the amblyopic eyes of patients with strabismic and anisometropic amblyopia compared to their non-amblyopic eyes or to the control group ( $p>0.05$ ). However, participants with anisometropic amblyopia had a thicker mean RNFL when compared to those with strabismic amblyopia ( $p>0.05$ ). Although there was a tendency for increased RNFL thickness across all quadrants in individuals with anisometropic amblyopia, this difference did not reach statistical significance ( $p<0.05$ ).

**Conclusion:** RNFL thickness was found to be similar in amblyopic and normal eyes. While existing studies have highlighted the impact of amblyopia on the RNFL, further investigations that consider additional factors are warranted.

**Keywords:** Anisometropia, amblyopia, retinal nerve fiber layer, strabismus

### Öz

**Amaç:** Bu çalışmanın amacı, şaşılık ambliyopisi ve anizometropik ambliyopisi olan hastaların ambliyopik gözleri ile ambliyopi olmayan gözleri ve kontrol grubundaki sağlam gözler arasında retina sinir lifi tabakası (RSLT) kalınlığında farklılık olup olmadığını optik koherens tomografi (OKT) kullanarak araştırmaktır.

**Yöntem:** Kırk, tek taraflı şaşılık ambliyopisi olan, 36 tek taraflı anizometropik ambliyopi ve 42 sağlıklı birey çalışmaya dahil edildi. Tüm olguların öykü, ortalama ve kadrant RSLT kalınlığı değerleri OKT kullanılarak kaydedildi.

**Bulgular:** Çalışmaya 64'ü kadın, 54'ü erkek olmak üzere toplam 118 kişi dahil edildi. Ortalama yaş 16,25±9,05 yılı. Tek taraflı ambliyopili olguların ambliyopili gözlerinin ortalama ve kadrant RSLT kalınlığı değerleri ambliyopisi olmayan gözlerle ve kontrol grubunun sağ gözleriyle karşılaştırıldığında istatistiksel olarak anlamlı bir fark gözlenmedi ( $p>0,05$ ). Hem şaşılık hem de anizometropik ambliyopili olguların ambliyopili gözleri arasında, ambliyopisi olmayan gözlerle veya kontrol grubunun sağ gözlerine göre ortalama ve kadrant RSLT kalınlığı açısından anlamlı bir fark yoktu ( $p>0,05$ ). Şaşılık ambliyopili olguların ambliyopik gözleri anizometropik ambliyopili olgularla karşılaştırıldığında, anizometropik ambliyopili olgularda anlamlı derecede daha kalın ortalama RSLT kalınlığı gözlemlendi ( $p>0,05$ ). Anizometropik ambliyopili olgularda şaşılık ambliyopili olgularla karşılaştırıldığında dört kadranda da RSLT'nin kalınlaşması yönünde bir eğilim olmasına rağmen bu fark istatistiksel olarak anlamlı değildi ( $p<0,05$ ).

**Sonuç:** Çalışmamızın sonuçları, ambliyopik ve sağlıklı gözler arasında RSLT kalınlığı açısından anlamlı bir fark olmadığını göstermektedir. Şu ana kadar yapılan çalışmalar ambliyopinin RSLT üzerindeki etkilerini göstermiştir. Ancak bu alanda farklı faktörlerin dikkate alındığı daha fazla araştırmaya ihtiyaç vardır.

**Anahtar kelimeler:** Ambliyopi, anizometropi, retina sinir lifi tabakası, şaşılık



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## Introduction

Amblyopia is a condition characterized by significantly reduced visual acuity, which may result from severe visual deprivation or abnormal binocular interactions during visual development (1). The prevalence of amblyopia ranges from 1% to 5% in studies of large populations (2-6). Amblyopia is not solely an eye-related issue; it can also be considered a form of brain dysfunction caused by exposure to abnormal visual stimuli. In amblyopic eyes, a decrease in contrast sensitivity, distortion of object shapes, and diminished spatial localization perception are commonly observed (7). There are three primary causes of amblyopia: Anisometropic amblyopia, strabismic amblyopia, and stimulus deprivation amblyopia. Anisometropic amblyopia may occur independently or in conjunction with strabismic amblyopia (8). Optical coherence tomography (OCT) facilitates the detailed evaluation of retinal structures, including retinal nerve fiber thickness (RNFL), macular volume, and macular thickness. Advancements in OCT technology allow for an in-depth examination of retinal layers, RNFL, and the choroid layer (9). Studies investigating the relationship between amblyopia and RNFL thickness have yielded mixed results. While some researchers found no significant differences in RNFL thickness between amblyopic and healthy eyes (10,11), others reported notable variations in RNFL thickness in anisometropic amblyopic eyes compared to normal eyes (12,13).

The objective of this study is to determine whether RNFL thickness varies between amblyopic eyes and non-amblyopic eyes in patients diagnosed with strabismic and anisometropic amblyopia, using OCT technology.

## Materials and Methods

This study was designed retrospective and cross-sectional. For the study, 40 strabismic patients with unilateral amblyopia, 36 anisometropic patients with unilateral amblyopia, and 42 healthy subjects who were admitted to İstanbul Okmeydanı Training and Research Hospital between January 2008 and January 2013 were included in the study. Approval was obtained from the İstanbul Okmeydanı Training and Research Hospital Ethics Committee with the decision dated 07/06/2013 and numbered 89, following the rules of the Declaration of Helsinki.

Inclusion criteria were as follows:

- Best-corrected visual acuity  $\geq 20/20$  in the better eye.
- Age range: 7 to 40 years.

- Intraocular pressure (IOP)  $< 20$  mmHg in both eyes.
- Clear ocular environment.
- Normal fundus examination.
- Unilateral amblyopia due to strabismus or anisometropia.

Exclusion criteria were as follows:

Subjects with organic eye disease; a history of intraocular surgery; opacity in the optic axis; a history of neurological disease; any retinal or optic nerve pathology; a history of glaucoma; IOP higher than 21 mmHg; nystagmus; non-communicable diseases; and age less than seven years were excluded.

Based on these criteria, the participants were divided into three groups according to the type of amblyopia:

- Group I: This group included forty patients with strabismic amblyopia and a shift of more than ten prism diopters.
- Group II: Thirty-six patients with hypermetropic anisometropic amblyopia and a refractive difference of at least two diopters between the two eyes were included in this group.
- Group III: Forty-two patients with complete visual acuity in both eyes and no ophthalmic or systemic disease were included in this group. The right eye values of the individuals were used in the study.

A detailed medical history was recorded, and uncorrected and best-corrected visual acuity, measured with the Snellen chart, and IOP, measured with a Topcon CT.80A pneumotonometer, were also recorded. The distance and near shifts of strabismic patients were recorded in prism diopters. Anterior segment and fundus examinations of all patients with fully dilated pupils were recorded. RNFL measurements were recorded by a single examiner using the Cirrus HD-OCT (model 4000, software version 5.1.1.6, Carl Zeiss Meditec, Inc.) with fully dilated pupils. Cases with an OCT signal strength of less than five were not included in the study. The average, superior, nasal, inferior, and temporal quadrant RNFL thicknesses were recorded in all cases.

Amblyopia cases were divided into three groups: mild, moderate, and severe in terms of best-corrected visual acuity measured from the Snellen chart by correcting the refractive error.

## Statistical Analysis

Data from all cases were entered into SPSS (Statistical Package for Social Science, Worldwide Headquarters SPSS

Inc.) 20.0 for statistical analysis. Means, standard deviations, proportions, and frequencies were used for descriptive statistics of the data. The Kolmogorov-Smirnov test was used to test the distribution of variables. ANOVA (Tukey test) and Kruskal-Wallis (Mann-Whitney U test) were used to analyze quantitative data. The paired sample t-test and the Wilcoxon test were used for repeated measurements. The chi-square test was used to analyze qualitative data.

## Results

### Demographics

A total of 118 cases were analyzed, with a mean age of  $16.25 \pm 9.05$  years (range: 7-40 years).

- Group 1 (strabismic amblyopia): Mean age  $14.05 \pm 6.89$  years (range: 7-27 years).
- Group 2 (anisometropic amblyopia): Mean age  $18.89 \pm 11.02$  years (range: 7-40 years).
- Group 3 (control): Mean age  $16.10 \pm 8.62$  years (range: 7-32 years).

Gender distribution:

- 64 participants (54.2%) were female.
- 54 participants (45.8%) were male.

There was no statistically significant difference in age or gender among the groups (Table 1).

When grouped according to visual acuity, in Group 1, 16 cases (40%) were classified as severe amblyopia, 14 cases (35%) as moderate amblyopia, and 10 cases (25%) as mild amblyopia. In Group 2, 16 cases (44.4%) were classified as severe amblyopia, 10 cases (27.8%) as moderate amblyopia, and 10 cases (27.8%) as mild amblyopia. No statistically significant difference in the degree of amblyopia was observed between Groups 1 and 2 based on visual acuity (Table 2).

### Overall RNFL Thickness

The mean RNFL thickness in the amblyopic eyes of 76 patients with unilateral amblyopia was  $95.81 \pm 13.33$   $\mu\text{m}$ , compared to  $94.92 \pm 13.94$   $\mu\text{m}$  in their normal eyes. This difference was not statistically significant ( $p > 0.05$ ).

RNFL thickness in the four quadrants for amblyopic versus non-amblyopic eyes:

- Temporal:  $65.50 \pm 21.18$   $\mu\text{m}$  vs.  $66.23 \pm 14.23$   $\mu\text{m}$ .
- Nasal:  $70.39 \pm 14.39$   $\mu\text{m}$  vs.  $68.97 \pm 13.93$   $\mu\text{m}$ .

- Superior:  $119.18 \pm 22.28$   $\mu\text{m}$  vs.  $119.89 \pm 22.28$   $\mu\text{m}$ .
- Inferior:  $126.60 \pm 20.28$   $\mu\text{m}$  vs.  $126.10 \pm 22.78$   $\mu\text{m}$ .

No statistically significant difference was found between the quadrants ( $p > 0.05$ ). Similarly, no significant disparity was observed when comparing amblyopic eyes with the control group (Table 3).

### Group-specific RNFL Findings

#### 1. Group 1 (strabismic amblyopia):

- Mean RNFL thickness:
- Amblyopic eyes:  $92.95 \pm 13.81$   $\mu\text{m}$ .
- Normal eyes:  $90.55 \pm 14.04$   $\mu\text{m}$ .
- Quadrant thicknesses in amblyopic vs. normal eyes:
- Temporal:  $63.80 \pm 13.21$   $\mu\text{m}$  vs.  $63.00 \pm 11.46$   $\mu\text{m}$ .
- Nasal:  $68.40 \pm 16.14$   $\mu\text{m}$  vs.  $64.25 \pm 12.82$   $\mu\text{m}$ .
- Superior:  $121.55 \pm 25.64$   $\mu\text{m}$  vs.  $114.05 \pm 22.90$   $\mu\text{m}$ .
- Inferior:  $124.60 \pm 22.69$   $\mu\text{m}$  vs.  $120.50 \pm 24.04$   $\mu\text{m}$ .

No significant differences were found between amblyopic and normal eyes, nor when compared to the control group ( $p > 0.05$ , Table 4).

#### 2. Group 2 (anisometropic amblyopia):

- Mean RNFL thickness:
- Amblyopic eyes:  $99.00 \pm 12.19$   $\mu\text{m}$ .
- Normal eyes:  $99.77 \pm 12.26$   $\mu\text{m}$ .
- Quadrant thicknesses in amblyopic vs. normal eyes:

**Table 1. The demographic characteristics of the cases**

|         | Age               | Sex    |      |      |      |
|---------|-------------------|--------|------|------|------|
|         |                   | Female |      | Male |      |
|         |                   | n      | %    | n    | %    |
| Group 1 | $14.05 \pm 6.89$  | 22     | 54.2 | 18   | 45.8 |
| Group 2 | $18.89 \pm 11.02$ | 20     | 55.6 | 16   | 44.4 |
| Group 3 | $16.10 \pm 8.62$  | 22     | 52.4 | 20   | 47.6 |
| p       | 0.104             | 0.955  |      |      |      |

**Table 2. Distribution of amblyopia severity within the groups**

| Visual acuity | Group 1 |      | Group 2 |      | p     |
|---------------|---------|------|---------|------|-------|
|               | n       | %    | n       | %    |       |
| Severe        | 16      | 40.0 | 16      | 44.4 | 0.796 |
| Moderate      | 14      | 35.0 | 10      | 27.8 |       |
| Mild          | 10      | 25.0 | 10      | 27.8 |       |

- Temporal:  $67.38 \pm 27.57 \mu\text{m}$  vs.  $69.83 \pm 16.20 \mu\text{m}$ .
- Nasal:  $72.61 \pm 11.98 \mu\text{m}$  vs.  $74.22 \pm 13.38 \mu\text{m}$ .
- Superior:  $122.50 \pm 18.14 \mu\text{m}$  vs.  $126.38 \pm 20.39 \mu\text{m}$ .
- Inferior:  $128.83 \pm 17.28 \mu\text{m}$  vs.  $132.33 \pm 19.79 \mu\text{m}$ .

There was no statistically significant difference between amblyopic and normal eyes and control group ( $p > 0.05$ , Table 4).

### Comparative Analysis Between Groups

- When comparing amblyopic eyes in Group 1 (strabismic amblyopia) and Group 2 (anisometropic amblyopia), anisometropic amblyopic eyes exhibited a significantly thicker mean RNFL ( $p < 0.05$ ).
- Although RNFL thickness in anisometropic amblyopic eyes appeared greater in all four quadrants, this variance was not statistically significant ( $p > 0.05$ , Table 4).

## Discussion

Amblyopia is known to cause many histologic changes in the LGN and visual cortex. In its pathogenesis, the balance of binocular competition is disturbed because the afferent pathways of the dominant eye stimulate more neurons in the visual cortex, resulting in decreased visual acuity in the non-dominant eye (14). RNFL thickness is affected by various parameters such as race, age, and sex. There may be many reasons for different results in different studies on this topic. These may include the use of different devices (e.g., OCT with a temporal analysis system or OCT with Fourier

principle), measurements performed by different clinicians, insufficient numbers of subjects enrolled in the study, or unequal gender distribution. In our study, there was no statistically significant disparity in terms of age and gender. After the detection of changes in the LGC and visual cortex, it has been a matter of curiosity whether amblyopia causes changes in the retina, and many studies have been conducted in this direction. A study conducted with 37 patients in 2009 found no statistically significant variance in the average and 4-quadrant RNFL thickness between amblyopic and healthy eyes. However, it was reported that the results could not be generalized to all amblyopes because the study did not involve patients with high degrees of amblyopia and patients with deprivation amblyopia (15). In our study, when amblyopic eyes (strabismic and anisometropic) were compared with normal eyes, no significant difference was observed between the mean RNFL thickness and 4-quadrant RNFL thicknesses. When the amblyopic eyes (strabismic and anisometropic) were compared with the right eye of the control group, no significant difference was observed in the mean RNFL thickness and in the thickness of the four quadrants. In our study, when the amblyopic eyes of strabismic and anisometropic amblyopes were compared, the RNFL of the anisometropic group was thicker than that of the strabismic group. This finding is consistent with those of both Repka et al. (15) and Kee et al. (16). Kavitha et al. (17) conducted a study utilizing the Fourier OCT in children and found that there was no discernible variance in RNFL thickness among patients with unilateral anisometropic amblyopia. Furthermore, they observed that there was no substantial alteration in RNFL thickness following one year

**Table 3. Average and quadrant thickness values of the amblyopia eye, fellow eye and control group**

| RNFL thickness | Amblyopia eye      | Fellow eye         | Control group      | P1    | P2    |
|----------------|--------------------|--------------------|--------------------|-------|-------|
| Mean           | $95.81 \pm 13.33$  | $94.92 \pm 13.94$  | $95.80 \pm 10.98$  | 0.470 | 0.998 |
| Temporal       | $65.50 \pm 21.18$  | $66.23 \pm 14.23$  | $67.66 \pm 11.30$  | 0.610 | 0.540 |
| Nasal          | $70.39 \pm 14.39$  | $68.97 \pm 13.93$  | $69.28 \pm 10.61$  | 0.377 | 0.663 |
| Superior       | $119.18 \pm 22.28$ | $119.89 \pm 22.28$ | $123.04 \pm 21.21$ | 0.790 | 0.361 |
| Inferior       | $126.60 \pm 20.28$ | $126.10 \pm 22.78$ | $124.66 \pm 13.85$ | 0.824 | 0.582 |

P1: Amblyopia and fellow eye, P2: Amblyopia and control group, RNFL: Retinal nerve fiber layer

**Table 4. Comparing the thickness of RSLT between groups**

| RNFL thickness | Group 1            | Group 2            | Group 3            | P1    | P2    |
|----------------|--------------------|--------------------|--------------------|-------|-------|
| Mean           | $92.95 \pm 13.81$  | $99.00 \pm 12.19$  | $95.80 \pm 10.98$  | 0.048 | 0.228 |
| Temporal       | $63.80 \pm 13.21$  | $67.38 \pm 27.57$  | $67.66 \pm 11.30$  | 0.465 | 0.953 |
| Nasal          | $68.40 \pm 16.14$  | $72.61 \pm 11.98$  | $69.28 \pm 10.61$  | 0.205 | 0.198 |
| Superior       | $114.05 \pm 22.90$ | $122.50 \pm 18.14$ | $123.04 \pm 21.21$ | 0.081 | 0.151 |
| Inferior       | $120.50 \pm 24.04$ | $128.83 \pm 17.28$ | $124.66 \pm 13.85$ | 0.090 | 0.241 |

P1: Group 1 and group 2, P2: Group 2 and group 3, RNFL: Retinal nerve fiber layer

of closure therapy (17). Huynh et al. (18) conducted a study using OCT temporal analysis and found no disparity in RNFL thickness among patients with unilateral amblyopia. In studies conducted in our country, similar to our study, Yazıcı et al. (19) compared the RNFL thickness of amblyopic and normal eyes of 114 patients, 67 of whom were strabismic, 35 of whom were anisometropic, and 12 of whom had deprivation amblyopia, using OCT with a temporal analysis system, and found no significant disparity in any quadrant. In the study conducted by Soyugelen et al. (20) using Fourier principle OCT in children aged 5-23 years, there was no difference between RNFL thicknesses. Similarly, Ulaş et al. (21) evaluated 32 unilateral anisometropic amblyopic patients with Fourier-domain OCT. They reported that the global, nasal, and inferonasal segments of the RNFL were thicker in anisometropic amblyopic eyes than in eyes with good vision. Contrary to the above studies, Soydan et al. (22) used OCT with a temporal analysis system in 50 unilateral anisometropic amblyopes and 50 normal subjects aged 5-62 years. The study revealed that the RNFL thickness was greater in anisometropic amblyopic cases compared with the control groups, and increased hyperopia and decreased axial length may have contributed to the difference between the groups (22). In the study by Yen et al. (12), the RNFL of 20 strabismic and 18 anisometropic amblyopic cases and 17 non-amblyopic anisometropic control cases was measured by OCT (Model 2000). The average RNFL of amblyopic eyes was determined to be significantly thicker than that of normal eyes. In line with the findings of this study, there was no significant variance in the RNFL between the amblyopic eyes of individuals with strabismic amblyopia and normal eyes, whereas those with anisometropic amblyopia exhibited a significantly thicker RNFL compared to normal eyes. Within the control group, no significant distinction in RNFL values was observed between the eye with high refractive error and its counterpart. In light of these findings, Yen et al. (12) explained the thick RNFL in amblyopic eyes with the theory that the postnatal decrease in ganglion cell numbers requires a sharper focus according to the stimulus. However, because this is not possible in amblyopic subjects, the necessary decrease does not occur. They also attributed the difference between the strabismic and anisometropic groups to the fact that neuronal loss in amblyopia is different depending on the etiology, and but suggested that new histopathologic studies were needed to confirm this (12). Yoon et al. (13) examined macular thickness and RNFL in 31 hypermetropic anisometropic amblyopic patients using OCT (Model 3000) and found that the mean RNFL was significantly thicker in amblyopic eyes ( $115.2 \pm 9.7 \mu\text{m}$ )

than in normal eyes ( $109.6 \pm 8.4 \mu\text{m}$ ). Again, based on these findings, they suggested that amblyopia has the potential to impact the RNFL while not affecting macular thickness. Other studies, including postmortem studies, should support this. In the literature, it has been emphasized that neural tissue loss may vary depending on the etiology of amblyopia (13). This is supported by the fact that in deprivation amblyopia and anisometropic amblyopia, there is a decrease in the LGC in monocular and binocularly innervated regions, whereas in strabismic amblyopia, there is a decrease only in binocularly innervated regions (23,24). Salchow et al. (25) showed that a refractive error of 1 diopter causes an increase in RNFL thickness of approximately  $1.67 \mu\text{m}$ . Therefore, we should consider that this increase in thickness in anisometropic cases may be due to the effect of refractive error on RNFL measurement. In a recent study, it was determined that the retinal nerve fiber layer (RNFL) of 35 eyes —comprising healthy, amblyopic, and single-vessel anisometropic eyes— between the ages of 6 and 63 years was examined using OCT. It was found that there was no significant difference between the RNFL studies of amblyopic eyes and the eyes of healthy subjects (26). In a study conducted in our country in 2022, the thickness of the RNFL in 111 pediatric patients with unilateral amblyopia due to anisometropia or strabismus, was investigated using OCT. It was found that the RNFL thickness of the superior, nasal, and temporal quadrants did not differ significantly. The study suggested that in amblyopic patients, there may be some damage in higher visual pathways such as the lateral geniculate nucleus and visual cortex rather than structural damage in the retina (27).

### Study Limitations

The limitation of this study is that the amblyopic patients' data were obtained from medical sources of our hospital. Another limitation is that the role of strabismus in the degree of amblyopia and retinal development may have affected the RNFL thickness measurements. However, the important feature of our study is that the groups are similar in terms of number, demographics, and long-term data collection.

### Conclusion

In our research, we found no substantial variance in the RNFL thickness of amblyopic and non-amblyopic eyes, both between amblyopic patients and between normal subjects. Studies on the effect of amblyopia on the RNFL have yielded different results. These differences may be due to the age factor, amblyopia type and severity, the insufficient

number of cases, and the position of the scanning ring used during OCT. Further research is needed in this area, taking into account different factors.

## Ethics

**Ethics Committee Approval:** Approval was obtained from the İstanbul Okmeydanı Training and Research Hospital Ethics Committee with the decision dated 07/06/2013 and numbered 89, following the rules of the Declaration of Helsinki.

**Informed Consent:** Retrospective study.

## Footnotes

### Authorship Contributions

Concept: M.A.Y., B.E., M.E., Design: M.A.Y., B.E., M.E., Data Collection or Processing: M.A.Y., B.E., M.E., Analysis or Interpretation: M.A.Y., B.E., M.E., Literature Search: M.A.Y., B.E., M.E., Writing: M.A.Y., B.E., M.E.

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