e-ISSN: 0975-1556, p-ISSN:2820-2643

Available online on www.ijpcr.com

International Journal of Pharmaceutical and Clinical Research 2023; 15(10); 674-679

Original Research Article

# Corneal Endothelial Cell Count, Corneal Thickness and Ageing: A Statistical Analysis in Indian Context and Comparison with Other Ethnic Groups

**Sanjiv Kumar[1], Malvika Dhar[2], Vivek Rajvanshi[3]**

[1]Professor, Department of Ophthalmology, Subharti Medical College, Meerut, U. P., India
[2]Assistant Professor, Department of Ophthalmology, Subharti Medical College, Meerut, U. P., India
[3]Associate Professor, Indian Institute of Management, Kolkata, W.B., India

**Received: 25-07-2023 / Revised: 28-08-2023 / Accepted: 30-09-2023**
**Corresponding author: Dr. Malvika Dhar**
**Conflict of interest: Nil**

**Abstract:**

**Aim:** This study aims to analyse patterns in endothelial cell count and corneal thickness in different age groups of Indian population and to compare findings with that of other ethnic groups.

**Materials and Methods:** This was a retrospective study of 7129 Indian subjects aged 11 years and above who had undergone specular microscopy and corneal endothelial cell count measurement at Subharti Eye Bank, Chatrapati Shivaji Subharti Hospital, affiliated with Swami Vivekanand Subharti University, Meerut (India). Information such as age, sex, eye, average cell density, average cell area, CV, maximum cell area and minimum cell area as measured by the specular microscope, were collected and analyzed. Data is analysed using STATA software.

**Observations and Results:** In this study a total of 7129 subjects above the age of 11 years were included out of which 3296 were male and 3833 were females. All subjects were divided into 7 age groups; 11-20 years, 21-30 years, 31-40 years, 41-50 years, 51-60 years, 61-70 years and more than 70 years. We found a negative correlation relation between mean corneal endothelial cell density (MCD) and age (correlation -0.2449, p-value 0.0000), and between mean corneal thickness (MCT) and age (correlation -0.1152, p-value 0.0000). The correlation analysis suggested that MCD and MCT decrease as the age advances. The mean cell area increases with advancing age (correlation +0.0928, p-value 0.000). The correlation between CV and age was positive (correlation = 0.0054, p-value=0.3242) but not statistically significant. Corneal endothelial cell characteristics in Indian population were compared with American, Japanese, Chinese, Malay, and Nigerian population. MCD was found to be highest in Japanese population in every age group and lowest in Indian population. However, MCD decreases with the advancing age in all the ethnic groups. MCD is higher in males than female population after the age of 30 years while it is higher in females of age less than 30 years. Rate of endothelial cell loss is more in males than females in the age group of 21-30 and 61-70 years while it is higher in females in the age groups of 31-40, 41-50, 51-60 and above 70 years of age. Cell loss rate is higher in Indian population as compared to Egyptian population in every age group.

**Conclusion:** The present study concluded that there is a statistically significant decline of MCD and MCT with age and also compared the results with other ethnic groups like Japanese, Chinese, American, Malaysian and Nigerian.

**Keywords:** Corneal endothelial cells, Specular microscopy, Mean cell density.



## Introduction

Corneal endothelial cells (CECs), which originate from neural crest, cover the posterior surface of the cornea. They comprise a monolayer of interdigitated cells arranged in a mosaic pattern that are mostly hexagonal in shape [1]. These cells are metabolically active and they are responsible for regulating fluid and solute transport between aqueous humor and corneal stroma in order to maintain normal corneal thickness and corneal transparency [2]. Corneal endothelial cell density is essential in the clinical assessment of cornea to determine its function and ability to respond to stress. It is important to determine a reference range for different population groups in order to establish abnormal values and aid predictability, especially with the noted rise in intraocular surgeries [3]. Unlike corneal epithelium, CECs cannot regenerate and decline throughout life [2,4]. In order to compensate for the cell loss, surrounding CECs enlarge, thus wound healing is

accomplished by spreading of cells forming a continuous layer of cells on the inner surface of the cornea [2]. Apart from endothelial cell density, the coefficient of variation (CV) of the mean cell area (Standard deviation of mean cell area/ Mean cell area) is a clinically valuable marker which is about 0.25 in the normal cornea. This increase in the variation of cell size is called as polymegathism.

Corneal thickness is another important parameter in the diagnosis of corneal disorders and treatment plan. Increased corneal thickness may give an artificially high IOP, while decreased corneal thickness causes an underestimation of IOP [5]. Corneal endothelium is metabolically active and utilises the Na+ K+ pump for keeping the stroma in a hydrated state of 70% water to prevent stromal oedema [6].

The average mean corneal endothelial cell density (MCD) is usually highest at birth (3000 cells/mm2) and gradually declines with age. A minimum of 400-500 cells/mm2 is required for the endothelial pump mechanism to function optimally. Values below this range are associated with corneal decompensation leading to Bullous Keratopathy [7]. Measurement of corneal endothelial cell count is of paramount importance as per literature. As specular microscopy is not easily accessible to all eye surgeons, it is expected that after this study they will get a gross idea regarding the average endothelial cell count in different age groups and both genders in the Indian population.

**Aims and Objectives**

1. To examine the relationship between corneal endothelial cell density (MCD) and age, and mean corneal thickness (MCT) and age of Indian population.
2. To compare the corneal endothelial cell density in Indian males and females.
3. To compare the corneal endothelial cell density in Indian population with that of other ethnic groups.

**Materials and Methods**

Ethical approval was obtained from the Ethical Committee of Subharti Medical College, Meerut. Patient consent was not required as this was a retrospective study. However, approval was obtained from the Head of Department of Ophthalmology, Subharti Hospital for retrieval of the necessary patient data. This was a retrospective study looking at the records of individuals who had undergone specular microscopy and corneal endothelial cell count measurement at Subharti Eye Bank, Chatrapati Shivaji Subharti Hospital, U.P. (India). Topcon model of specular microscope had been used for this study.

**Inclusion Criteria**

1. Individuals of 11 years of age and above
2. Individuals who are born in India

**Exclusion Criteria**

1. Individuals with a previous history of ocular trauma
2. Individuals with a history of ocular surface disease
3. Individuals with a history of ocular surgery or any endothelial dysfunction.

**Data Collection**

Data such as age, sex, eye, average cell density, standard deviation, average cell area, CV, maximum cell area and minimum cell area as measured by the specular microscope was retrieved and analysed.

In total we retrieved 7129 records, which is a very large sample as compared to other studies carried on the similar subject, therefore, results may be generalized. However, records in different age groups were not uniformly distributed.

**Data Analysis**

Data analysis was performed using STATA software. Unpaired t-tests were used to test the statistical significance in difference of the variables used in analysing the data.

Pearson correlation coefficient and linear regression analysis were conducted to examine the change in endothelial cell characteristics with age.

The t-test statistics have been provided within the tables shown and p-values at 1%, 5% and 10% are highlighted in the tables with ***, **, * to show the statistically significance of the analysis.

**Results**

**Table 1: Number of Male and female subjects in different age groups of Indian population**

| Age (Years) | Male No. | Female No. | Total |
|---|---|---|---|
| 11-20 | 24 | 9 | 33 |
| 21-30 | 26 | 23 | 49 |
| 31-40 | 131 | 214 | 345 |
| 41-50 | 473 | 733 | 1206 |
| 51-60 | 1062 | 1423 | 2485 |
| 61-70 | 1121 | 1116 | 2237 |
| >70 | 459 | 315 | 774 |
| Total | 3296 | 3833 | 7129 |

Out of a total of 7129 subjects, 3296 were male subjects and 3833 were females.

**Table 2a: Corneal endothelial cell characteristics in different age groups**

| Age (Years) | No. of Subjects | Mean Corneal Endoth. Cell Density ( Cell/mm2 mean ± SD) | CV in cell size (% mean ± SD) | Mean Cell area (mm2 mean ± SD) | Corneal Thickness |
|---|---|---|---|---|---|
| 11-20 | 33 | 2296.0±389.2 | 47.52±12.42 | 697.6±184.6 | 0.5187±.037 |
| 21-30 | 49 | 2107.7±314.2 | 54.22±11.52 | 780.1±173.5 | 0.5141±.033 |
| 31-40 | 345 | 2079.7±296.6 | 52.58±12.01 | 783.6±167.6 | 0.5117±.037 |
| 41-50 | 1206 | 2004.5±295.3 | 52.80±11.72 | 788.9±166.8 | 0.5050±.035 |
| 51-60 | 2485 | 1912.9±278.6 | 52.78±11.44 | 810.1±161.9 | 0.5010±.036 |
| 61-70 | 2237 | 1879.9±272.7 | 52.97±11.64 | 822.1±161.2 | 0.4984±.036 |
| >70 | 774 | 1841.2±251.9 | 52.65±11.68 | 823.7±158.5 | 0.4976±.037 |

The mean corneal endothelial cell density and mean corneal thickness decreases as the age advances, while the mean cell area increases with advancing age. To understand this relationship in a better way, we performed unpaired t-test and tested the statistical significance of the difference of MCD and MCT from the previous age group. Table 2b show the change in MCD and MCT from the last age group.

**Table 2b: Difference in MCD and MCT with Ageing**

| Age Group From : To | Mean Corneal Endoth. Cell Density ( Cell/mm2 mean ± SD | Corneal Thickness |
|---|---|---|
| 11-20 : 21-30 | -188.24*** (2.1446) | -0.0047 (0.5876) |
| 21-30 : 31-40 | -28.004 (0.6138) | -.0023 (0.4131) |
| 31-40 : 41-50 | -75.221*** (4.1671) | -0.0067*** (3.0430) |
| 41-51 : 51-60 | -91.6280*** (9.1862) | -0.0041*** (3.2057) |
| 51-60 : 61-70 | -32.97164*** (4.1005) | -0.0026*** (2.4101) |
| 61-70 : >70 | -38.6761*** (3.4660) | -0.0008 (0.5372) |

*, **, *** show level of significance at 10% (p-value < 0.1), 5% (p-value < 0.05), and 1% (p-value < 0.01), t-test statistics is reported in parenthesis.

It is clear from the Table 2b that MCD and MCT both decreases with the increase in age. Results for MCD were not significant when age increases from 21-30 to 31-40 age group, however the sign of MCD remains negative. The change in MCT with the growing age was significant between age 21 to 70.

Table-1 show that the sample size for age group 11-20, 21-30 and >70 years is smaller as compared to other age groups, there is a possibility that the concentration of the subjects may be around some particular ages and not uniformly divided in the representing age groups.

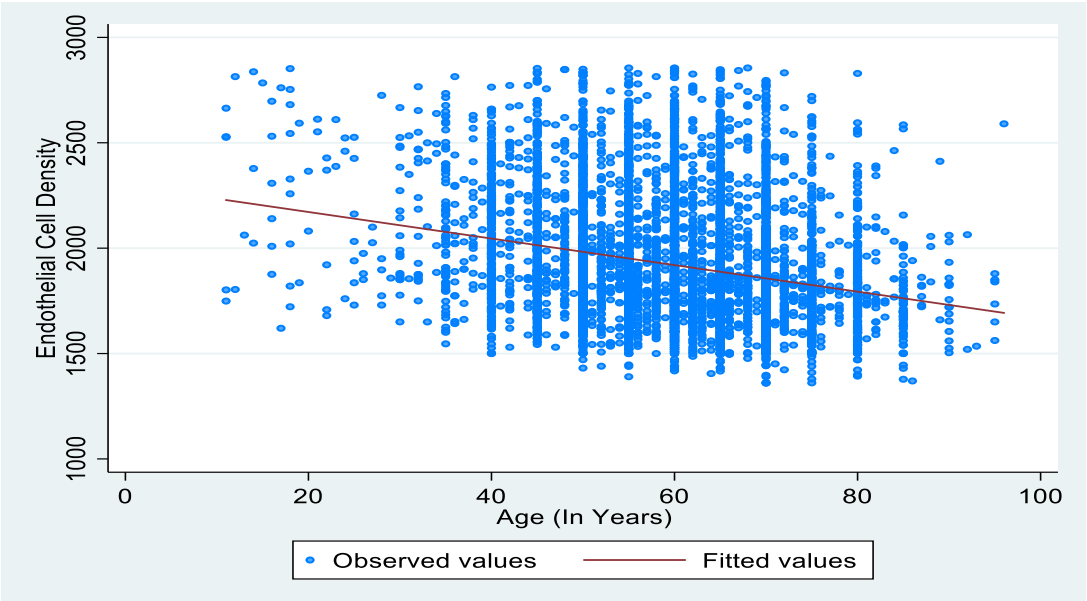


**Figure 1: Scatter Plot and Fitted Regression Line between MCD and Age**

Figure-1 shows scatter plot between observed value of MCD for all age. A regression line fitted (slope coefficient = - 6.299, p-value = 0.000) show that there is a negative linear relationship between MCD and age. For one-year increase in age MCD on average drops by 6.3.

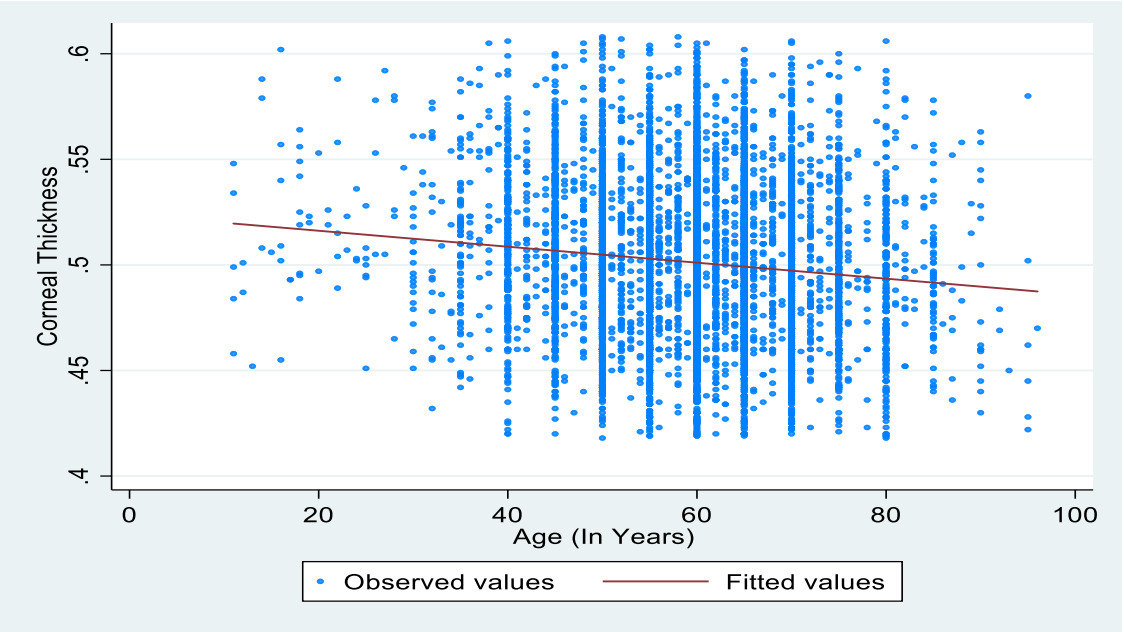


**Figure 2: Scatter Plot and Fitted Regression Line between MCT and Age**

Figure-2 shows scatter plot between observed value of MCT for all age. A regression line fitted (slope coefficient= - 0.0037, p-value = 0.000) show that there is a negative linear relationship between MCT and age. Results show that one year increase in age results in a drop of 0.0037 unit in MCT. Regression analysis confirm our findings that MCD and MCT decreases with ageing.

**Table 3: Comparison of mean corneal endothelial cell density in different population groups**

| Age group (years) | American [14] | | Japanese[14] | | Chinese[15] | | Malay[16] | | Nigerian[11] | | Indian | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | No of eyes | Cell density (cell/m$^2$; mean ± SD) | No of eyes | Cell density (cell/m$^2$; mean ± SD) | No of eyes | Cell density (cell/m$^2$; mean ± SD) | No of eyes | Cell density (cell/m$^2$; mean ± SD) | No of eyes | Cell density (cell/m$^2$; mean ± SD) | No of eyes | Cell density (cell/m$^2$; mean ± SD) |
| 11-20 | - | - | - | - | - | - | - | - | - | - | - | - |
| 21–30 | 11 | 2,977±324 | 18 | 3,893±259 | 100 | 2,988±243 | 49 | 2,783±286 | 81 | 2,860±227 | 49 | 2107±314 |
| 31–40 | 6 | 2,739±208 | 10 | 3,688±245 | 100 | 2,920±325 | 9 | 2,551±319 | 62 | 2,631±394 | 345 | 2079±296 |
| 41–50 | 11 | 2,619±321 | 10 | 3,749±407 | 97 | 2,935±285 | 10 | 2,744±239 | 31 | 2,433±442 | 1206 | 2004±295 |
| 51–60 | 13 | 2,625±172 | 10 | 3,386±455 | 97 | 2,810±321 | 15 | 2,509±228 | 37 | 2,545±319 | 2485 | 1912±278 |
| 61–70 | 8 | 2,684±384 | 6 | 3,307±330 | 90 | 2,739±316 | 25 | 2,560±304 | 81 | 2,538±362 | 2237 | 1879±272 |
| >70 | 15 | 2,431±339 | 5 | 3,289±313 | 83 | 2,778±365 | 17 | 2,570±301 | 67 | 2,493±353 | 774 | 1841±251 |

MCD is highest in Japanese population in every age group and lowest in Indian population. MCD decreases with the advancing age in all the ethnic groups.

**Table 4: Average corneal Endothelial Cell Density in Male & Female**

| Age ( Years) | Male Cell Density (cells/ mm2 mean±SD) | Female Cell Density (cells/ mm2 mean±SD) | Difference (Male – Female) |
|---|---|---|---|
| 11-20 | 2292.96 | 2304.11 | -11.15 (-.0722) |
| 21-30 | 2047.42 | 2175.96 | -128.53 (-1.4449) |
| 31-40 | 2091.12 | 2072.79 | 18.3201 (0.5561) |
| 41-50 | 2029.57 | 1988.37 | 41.1976*** (2.3694) |
| 51-60 | 1945.11 | 1888.87 | 56.2389*** (5.0011) |
| 61-70 | 1896.03 | 1863.75 | 32.2776*** (2.8026) |
| >70 | 1859.44 | 1814.75 | 44.6984*** (2.4326) |

*, **, *** show level of significance at 10% (p-value < 0.1), 5% (p-value < 0.05), and 1% (p-value < 0.01), t-test statistics is reported in parenthesis. MCD is higher in males than female population after the age of 30 years while it is higher in females of age less than 30 years. MCD decreases with the advancing age in both male and female subjects.

**Table 5: Cells loss rate according to increasing age in every 10 years**

| Age (Years) | Male | Female |
|---|---|---|
| 11-20 | -- | -- |
| 21-30 | 10.7% | 5.6% |
| 31-40 | 2.1% | 4.7% |
| 41-50 | 2.9% | 4.1% |
| 51-60 | 4.2% | 5.0% |
| 61-70 | 2.5% | 1.3% |
| >70 | 1.9% | 2.6% |

Rate of cell loss is more in males than females in the age group of 21-30 and 61-70 years while it is higher in females in the age groups of 31-40, 41-50, 51-60 and above 70 years of age.

**Table 6: Comparison of rate of cell loss in Indian population versus Egyptian population**

| Age (Years) | Male | Female | Egyptian population |
|---|---|---|---|
| 11-20 | -- | -- | |
| 21-30 | 10.7% | 5.6% | |
| 31-40 | 2.1% | 4.7% | 0.7% |
| 41-50 | 2.9% | 4.1% | 0.1% |
| 51-60 | 4.2% | 5.0% | 0.3% |
| 61-70 | 2.5% | 1.3% | 0.1% |
| >70 | 1.9% | 2.6% | 0.3% |

Table-6 compares the rate of endothelial cell loss in Indian population with that of Egyptian peers which shows that cell loss rate is higher in Indian population in every age group [12].

**Discussion**

Age related changes in corneal endothelial cell characteristics of various populations have been described in the literature. In the present study, we evaluated the corneal endothelial cell density and morphology in 7129 Indian subjects and compared them with other ethnic groups. The results revealed that MCD decreases as the age advances (r= -0.2449, p value 0.0000). A comparison with previous studies shows that MCD decreases with advancing age in Iranian eyes [9] (p<0.001, r=-0.64), Turkish eyes [10] (p<0.001, r=-0.388), Egyptian eyes [12] (p<0.001, r=-0.357) and Thai eyes [13] (p<0.001, r=-0.496). Populations of different ethnicities were divided into 7 age groups (11-20 years, 21-30 years, 31-40 years, 41-50 years, 51-60 years, 61-70 years and >70 years). On comparing MCD for every age group (table 3), it was observed that MCD was highest in Japanese population in every age group, followed by Chinese population while it was lowest in Indian population in all the age groups.

In our study, 3296 subjects were males and 3833 were females. MCD was compared in every age group of both the genders. Results showed that after the age of 40 years, MCD is higher in males than females (p< 0.01) while in other studies done on Turkish [11], Nigerian [10] and Egyptian [12] population, no statistically significant difference between genders was found.

In our study, MCT (Mean Corneal Thickness) has a negative correlation coefficient with age (r=-0.1152, p value 0.0000) which denotes that MCT decreases with advancing age, which is comparable with the study done on Egyptian population [12] (r=-0.113, p value 0.007) and Thai population [13] (r=-0.215, p value <0.001). MCA( Mean Cell Area) has a positive correlation with age in our study (r=+0.0928, p value 0.0000) which is also true for another study conducted in the year 2000 by SK

Rao, P Ranjan Sen, R Fogla et al on 537 Indian subjects where MCA shows a statistically significant increase in age (r=+0.362, p value <0.001) 8. Other studies done on Iranian [9] (r=+0.56, p value <0.001), Turkish [10] (r=+0.363, p value <0.001) and Egyptian eyes [12] (r=+0.111, p value =0.008) also showed a positive correlation of MCA with age.

In our study done on 7129 subjects, the correlation between CV and age was positive (correlation = 0.0054, p-value=0.3242) but not significant while in another study done on 537 Indian eyes by S K Rao there was a statistically significant increase in CV (correlation 0.096, p value = 0.02) [8]. According to our study, rate of endothelial cell loss is more in males than females in the age group of 21-30 and 61-70 years while it is higher in females in the age groups of 31-40, 41-50, 51-60 and above 70 years of age which is higher than that of Egyptian population [12]. (Table 5 and Table 6).

## Conclusion

We concluded that there is a statistically significant decline of MCD and MCT with age. We also compared our results with other ethnic groups like Japanese, Chinese, American, Malaysian and Nigerian. In the present scenario, enucleation of eye is rarely done. Only excision of corneoscleral button is carried out for corneal transplantation. The cost of specular microscope to evaluate the corneal endothelial cell characteristics is very high (approx. INR 35 lakh, approx. USD 50,000), We believe that this study will be useful to the eye banks as it provides an idea of the corneal endothelial cell characteristics of Indian population. The findings in this study can also be used as a baseline value for comparison between Indian and other ethnic groups helpful for intraocular surgeries and corneal transplantation.

## References


1. Waring GO 3rd, Bourne WM, Edelhauser HF et al. The corneal endothelium. Normal and pathologic structure and function. Ophthalmology. 1982; 89:531–90.
2. Mishima S. Clinical investigations on the corneal endothelium-XXXVIII Edward Jackson memorial lecture. Am J Ophthalmol. 1982; 93:1–29.
3. Kanski JJ, Bowling B, editors. Clinical Ophthalmology A Systematic Approach. Seventh ed. Philadelphia, PA: Elsevier; 2011. Chapter 6; p. 168
4. Bourne WM, McLaren JW. Clinical responses of the corneal endothelium. Exp Eye Res. 2004; 78:561–572. doi: 10.1016/ j.exer. 2003.08.002
5. Nishida T: Cornea. In: Krachmer J, ed. Cornea Vol. 1 Fundamentals of cornea and external disease. St. Louis: Mosby, 1997:17–19.
6. Doughty MJ, Zaman ML. Human corneal thickness and its impact on intraocular pressure measures: a review and meta-analysis approach. SurvOphthalmol. 2000; 44:367-408. doi: 10.1016/S0039-6257(00)00110-7.
7. Kirk RW. External Disease and Cornea. San Francisco, CA: American Academy of Ophthalmology; 2005. Basic and Clinical Science Course Section 8 2004–2005; pp. 32–33.
8. SK Rao, P Ranjan Sen, R Fogla et al. Corneal endothelial cell density and morphology in normal Indian eyes. Cornea. 2000; 19(6): 820-3.
9. Mohammad Naseer Hashemian, Sasan Moghimi, Masood AghsaieFard et al. Corneal endothelial cell density and morphology in normal Iranian eyes. BMC Ophthalmol. 2006; 6: 9.
10. Ceyhun Arici, Osman Sevki Arslan and Funda Dikkaya. Corneal endothelial cell density and morphology in healthy Turkish eyes. J Ophthalmol. 2014.
11. Temitope Ewete, Efeoghene Uchenna Ani and Adegbovega Sunday Alabi. Normal corneal endothelial cell density in Nigerians. Clin Ophthalmol. 2016; 10: 497-501.
12. Marwa Mahmoud Abdellah, Hatem Gamal Ammar, Mohamed Anbar et al. Corneal Endothelial cell density and morphology in healthy Egyptian eyes. J Ophthalmol. 2019.
13. Napaporn Tananuvat and Natawan Khumchoo. Corneal thickness and endothelial morphology in normal Thai eyes. BMC Ophthalmol. 2020; 20:167.
14. Matsuda M, Yee RW, Edelhauser HF. Comparison of corneal endothelium in an American and a Japanese population. Arch Ophthalmol. 1985; 103:68-70.
15. Yunliang S, Yuqiang H, Ying-peng L, Ming-zhi Z, Lam DSC, Rao SK. Corneal endothelial cell density and morphology in healthy Chinese eyes. Cornea. 2007; 26:130–132.
16. Mohammed-Salih PA. Corneal endothelial cell density and morphology in normal Malay eyes. Med J Malaysia. 2011; 66(4):300–303.