The Open Ophthalmology Journal

Content list available at: https://openophthalmologyjournal.com

RESEARCH ARTICLE

Age-related Changes of the Corneal Endothelium in the Hispanic Elderly Population

Jorge E. Valdez-García[1,*], Gustavo Ortiz-Morales[1,#], Nallely Morales-Mancillas[1], Jorge Luis Domene-Hickman[2], Julio Hernández-Camarena[1], Denise Loya-García[1], Judith Zavala[2] and Alejandro Rodriguez-García.[1]

[1]*School of Medicine & Health Sciences, Institute of Ophthalmology & Visual Sciences, Tecnologico de Monterrey, Monterrey, Mexico*
[2]*School of Medicine & Health Sciences, Tecnologico de Monterrey, Monterrey, Mexico*

Abstract:

Purpose:

To assess the corneal endothelial morphometry in healthy elderly Hispanic eyes.

Methods:

A retrospective, cross-sectional, observational, descriptive, and analytical study was designed to assess the central region of the corneal endothelium by specular microscopy. A total of 241 eyes from 125 patients were studied. All eyes included were from healthy patients 65 or older (mean: 74.56± 6.74), without ocular disease or surgeries.

Results:

The mean cell density (MCD) was 2198.92±493.43 cells/mm^2. 125 (51.87%) eyes had polymegethism, with a mean coefficient of variation (CV) of 42.89 ± 9.16%. 148 (61.41%) eyes presented pleomorphism, with an average of 46.25 ± 7.13% hexagonal cells. The mean corneal thickness was 527.55±28.37 µm. A statistically significant difference between age groups was found for MCD and pachymetry (P<0.05). 41 eyes (17.01%) had cornea guttata.

Conclusion:

This study suggests that the elderly Hispanic population has a high prevalence of polymegethism, pleomorphism, and guttata. Surprisingly, the oldest age group (>85) had an above-average MCD, with lower rates of polymegethism and pleomorphism.

Trial Registration Number: #ARCC/ECH2019

Date of Registration: October 18, 2019

Keywords: Cornea, Specular microscopy, Cell density, Pleomorphism, Polymegethism, Cornea guttata.

Article History	Received: November 18, 2021	Revised: January 3, 2022	Accepted: January 19, 2022

1. INTRODUCTION

The corneal endothelium is an essential part of the cornea's structure and function, playing a key role in maintaining corneal hydration homeostasis and transparency.

The measurement of central corneal thickness (CCT), endothelial cell density, size, and morphology is essential for evaluating the endothelial cell function for diagnostic purposes and the preoperative evaluation of any anterior segment surgery [1]. The total number of endothelial cells may be affected by aging, trauma, UV exposure, inflammation, surgery, *etc* [2 - 4].

Corneal endothelial cell alterations may affect corneal transparency, worsening visual outcomes. Age plays an essential role in this process since it has demonstrated a direct

* Address correspondence to this author at the School of Medicine & Health Sciences, Institute of Ophthalmology & Visual Sciences, Monterrey, Mexico, Av. Batallón de San Patricio No. 112. Col. Real de San Agustin. San Pedro Garza Garcia, N.L. C.P. 62770. México; Tel: +52 81 8888-0552;
Fax: +52 81 8356-1799; E-mail: jorge.valdez@tec.mx
# *These authors contributed equally*

correlation with endothelial cell loss by reducing pump functionality and corneal wound-healing, which contribute to reduced visual acuity [3, 5].

Various studies analyzing the corneal endothelial cell density in different populations highlight the importance of including the endothelial cell morphology in relation to ethnicity and age. Available data from these studies show statistically significant differences in endothelial cell parameters among ethnic groups and sexes [1, 6 - 13].

The differences in endothelial cell morphology among ethnic groups, age, and sex show that normative data on the endothelial cell parameters may help assess the endothelial cell function in individual patients and further our understanding of corneal endothelial diseases.

The present study aims to describe the cell density and morphological characteristics of the corneal endothelium of the healthy elderly Hispanic population.

2. SUBJECTS AND METHODS

A retrospective, cross-sectional, observational, descriptive, and analytical study was designed to study the specular microscopy characteristics of the corneal endothelium in elderly Hispanic patients. The study was conducted at the Tecnologico de Monterrey Institute of Ophthalmology and Visual Sciences between 2013 and 2019. All patients enrolled in the study participated voluntarily after reading and signing a written informed consent, previously reviewed and approved by our institution's Ethics and Research Committees (#ARCC/ECH2019), following the tenets of the Declaration of Helsinki.

The inclusion criteria comprised Hispanic patients over 65 years of age, living in Northern Mexico who came for a routine ophthalmological examination and were otherwise healthy. The exclusion criteria included a past medical history of ocular trauma, corneal or intraocular inflammation, the use of systemic drugs that could affect endothelial cells, evidence of endothelial disease during a slit-lamp examination, previous ocular surgery (including cataract surgery), increased intraocular pressure, corneal opacity, or retinal photocoagulation procedures.

After a complete medical history and general ophthalmological examination, all eyes of patients included in the study had specular microscopy (SM) performed by the same technician (SIS) using the EM-3000 specular microscope (Tomey®, Phoenix, AZ, USA) at a similar room temperature and humidity conditions. EM-3000 is a non-contact specular microscope that uses a charge-coupled device camera to capture 15 images in a series; it automatically selects and displays the finest among these images. The in-built software allows for fast and precise analysis of endothelial parameters. A wide range capturing area of 0.25 x 0.54 mm allows a larger count of cells. Corneal thickness measurement accuracy is +/- 10 μm.

Only the central corneal measurement point was considered for analysis. Parameters studied included the mean endothelial cell density (MCD), the mean cell area (MCA), the coefficient of variation (CV) of cell area (calculated as SD/mean cell area in μm), and the percentage of hexagonal cells (PHC), and the CCT. Polymegethism was considered a CV over 40% and pleomorphism when less than 50% of the endothelial cells showed hexagonality. SM images were classified as healthy when minor dark spots were present or as having guttae, mound-shaped excrescences in the Descemet's membrane.

Patients were classified according to age into five groups: 65-69, 70-74, 75-79, 80-84, and ≥85 years old. All statistical analyses were performed using STATA v. 12.0 (StataCorp® LLC, Texas, USA). Continuous data are presented in mean ± SD and categorical data as proportions. Since the data showed an abnormal distribution according to the Shapiro-Wilk test and distribution plots, Mann-Whitney U-test was performed to compare cell parameters according to gender and Kruskal-Wallis one-way analysis of variance to compare morphometry parameters between age groups. A multiple regression analysis was conducted to determine endothelial cell density changes with age, CV, MCA, PHC, and CCT. A *P* value less than 0.05 was considered statistically significant.

3. RESULTS

A total of 241 eyes from 125 patients corresponding to 64 (51.20%) males and 61 (48.80%) females over 65 years old, were studied. Nine pairs of eyes from nine patients had undergone previous cataract surgery and were therefore excluded for statistical analysis. A mean number of 219.46± 66.1 endothelial cells were counted. The endothelial cell density and morphology according to patients' age distribution are shown in Table **1**. Fig. (**1**) shows a collage of morphometrical parameters in relation to age.

Table 1. Endothelial cell density and morphology according to age.

Age group (y)	No. of eyes	Mean age (y)	MCD (cells/mm^2)	MCA (μm^2)	MCV (%)	Hexagonality (%)	MCCT(μm)	Mean no. of cells
65-69	63	66.8±1.5	2263.2±509.9	417.6±110.8	43.9±9.1	45.5±7.5	520.2±25.3	226.7
70-74	77	71.7+1.4	2092.4±487.8	454.8±157.3	43.2±10	46.3±7.3	530.2±26.1	209.4
75-79	35	77.4+1.5	2134.6±466.4	454.1±166.3	41.5±5.9	44.6±6.5	535.6±34	216.3
80-84	39	82.1±1.6	2260.2± 522.9	426.2±154.5	42.5±10.4	47.1±7.7	521.5±30.8	234.3
85-90	27	85.9±1.1	2347.2±412.1	412.4±88.7	41.9±8.3	48.5±4.9	535.1±25.6	210.3

MCD: Mean cell density, **MCA:** Mean cell area, **MCV:** Mean coefficient of variation, **MCCT:** Mean central corneal thickness.

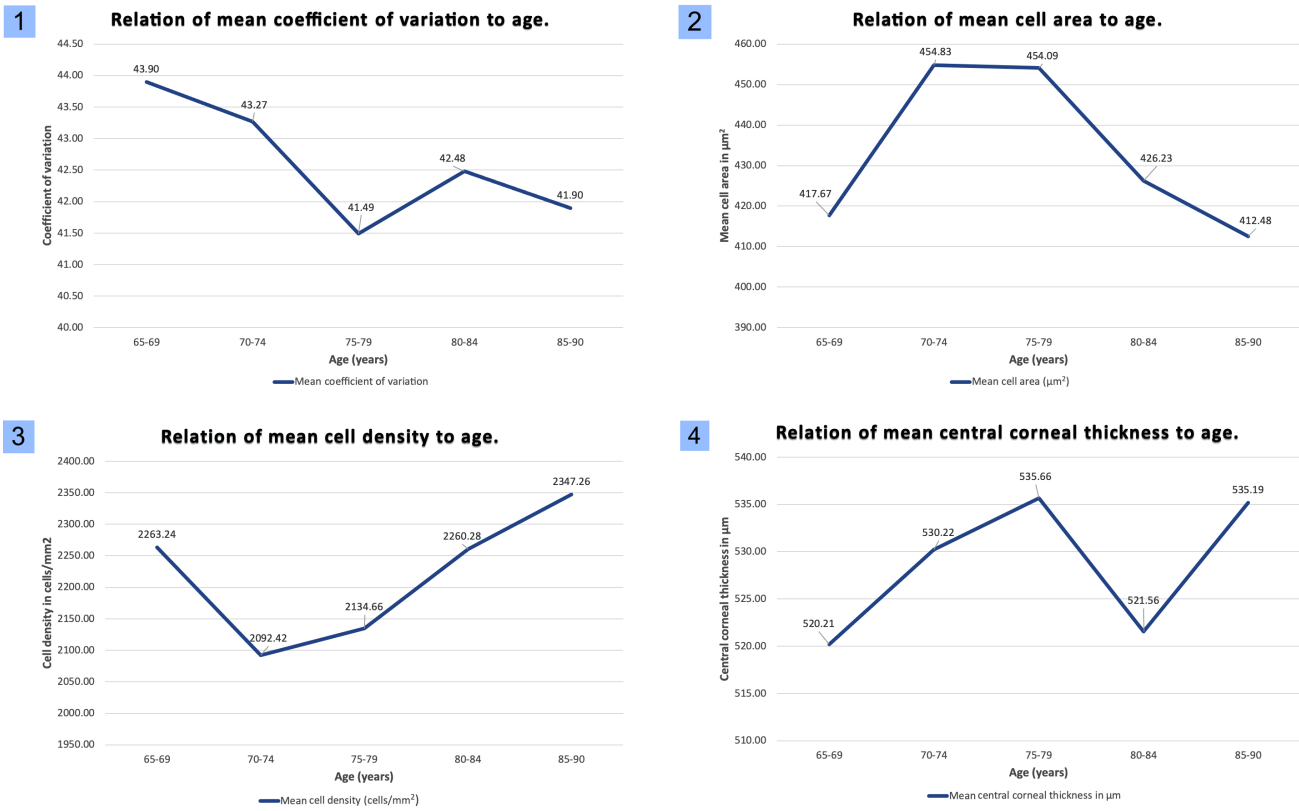

Fig. (1). A collage of morphometrical parameters in relation to age.

3.1. Mean Cell Density

The MCD across all age groups was 2198.92±493.43 cells/mm^2 (95% CI 2137 to 2261). A small decline was observed as age increased, except for the ≥85 years group, where surprisingly, an increase in cell density was observed (MCD: 2347.2 ± 412.1). The changes in MCD among age groups were statistically significant (P=0.003). MCD was higher in males than in females (2275.7 *vs*. 2120.1 cells/mm^2, P= 0.02). Fifteen eyes (6.22%) had a CD over 2800 cells/mm^2, while thirty (12.25%) had less than 1500 cells/mm^2; of those, only five (2.07%) had fewer than 1000 cells/mm^2.

3.2. Mean Cell Area

The MCA across all age groups was 435.63 ± 141µm^2 (95% CI 417.8 to 453.4). No statistically significant difference in MCA was observed among age groups (P=0.24).

3.3. Mean Coefficient of Variation

The MCV for all age groups was 42.89 ± 9.16% (95% CI 41.73 to 44.05). No statistical difference was observed among groups for MCV (P=0.73). The 65-69 years age group had the highest prevalence of polymegethism, while the 80-84 years age group had the lowest (60.32% *vs*. 38.4%, respectively).

3.4. Mean Hexagonality

The PHC was 46.25 ± 7.13% (95% CI 45.35 to 47.15). No statistical difference was observed among groups for PHC (P= 0.09). The 75-79 years age group had the lowest mean PHC, while the ≥85 years group had the highest (44.63% *vs*. 48.48%, respectively). Age was not a risk factor for pleomorphism (R=0.005, p=0.27) in this population. The 75-79 years age group had the highest prevalence of pleomorphism (77.14%), while the ≥85 years age group had the lowest (44.4%).

3.5. Mean Central Corneal Thickness

The mean CCT for all age groups was 527.55 ± 28.37µm (95% CI 524 to 531.1). A steady increase in CCT was observed in relation to age (R=0.012, *P*=0.09). A statistically significant difference was observed among age groups for the mean CCT (P=0.02). The lowest mean CCT value was recognized in the 65-69 years age group (520.2 ± 25.3µm) and the highest in the 75-79 years age group (535.6 ± 34µm)

Finally, a total of 41 eyes (17%) showed cornea guttata. A multiple regression analysis was run to predict cell density based on age, CV, MCA, PHC, and CCT: F (5,69) = 154.29, p= <0.0001, R^2= 0.9179. Variables, CV, MCA, and PHC added statistical significance to the prediction (p=<0.05).

4. DISCUSSION

Corneal endothelial cells are essential for maintaining corneal transparency by keeping the corneal stroma in a state of partial dehydration. Such homeostatic status is accomplished by a delicate balance between the rate of water entering the hydrophilic stroma through the endothelial barrier and its active removal by the action of the metabolic Na/K ATPase endothelial pump [14].

Table 2. Endothelial cell parameters and central corneal thickness by gender.

Parameter	Female	Male	P-value
Mean age (y)	74.43±6.39	74.68±7.09	0.90
MCD (cells/mm^2)	2120.1±519.7	2275.75±455.4	0.02
MCA (μm^2)	454±159.60	417.69±118.1	0.24
MCV (%)	43.91±10.21	41.9±7.92	0.08
Hexagonality (%)	45.21±7.92	47.2±6.14	0.08
MCCT(μm)	529±29.68	526.1±27.1	0.41

MCD: Mean cell density, **MCA:** Mean cell area, **MCV:** Mean coefficient of variation, **MCCT:** Mean central corneal thickness.

Table 3. The average endothelial cell density (cells/mm2) of healthy eyes among various populations.

Population	Author	MCD (cells/mm^2)	Age (y)	No. of eyes
Lithuania	Galgauskas[1]	2366	60 – 89	158
Philippines	Padilla[6]	2780.45	61 – 86	200
India	Rao[9]	2404	60 – 87	142
Turkey	Goktas[10]	2,215	61 – 90	371
Iran	Hashemian[11]	1681.61	61 – 85	225
Central Mexico	Graue[12]	1805.33	60 – 89	25
Central Mexico	Molina[13]	1910	60 – 89	50
North Mexico	Valdez *et al*.	2198	65 – 90	241

MCD: Mean cell density.

After birth, the corneal endothelial cell density (ECD) peaks at approximately 6000 cells/mm^2 which starts to decline during the first years of life due to corneal growth [15]. Then, cell density continues its gradual decay over time, particularly between the ages of 20 to 80 years at an estimated annual rate of 0.6% [5]. This slowly progressive cell loss is accompanied by a proportional increase in polymorphism and polymegethism [16].

When aging, toxicity, trauma, inflammation, or disease produce an ECD drop below 500 cells/mm^2, irreversible stromal edema occurs, and the cornea eventually loses transparency [17]. For many years it was thought that corneal endothelial cells could not regenerate. However, recent investigations have shown that endothelial cells retain their capacity to divide and renew, although they rarely do so [18]. A hypothesis that may help explain, at least in part, such cellular behavior postulates that certain groups of dividing cells may enter a process of cellular senescence or replicative failure that limits their capacity to maintain their lifespan [19, 20]. This hypothesis is supported by an age-related increase in the number of senescent cells in the human corneal endothelium [21, 22]. Despite the relentless pursuit of a better understanding of the biologic mechanisms implicated in the gradual endothelial cell loss found during aging, there are still more questions than answers. This phenomenon is probably multifactorial, involving environmental, hormonal, biochemical, and physiological processes related to aging [23].

Several studies from diverse populations worldwide analyze the corneal endothelial cell density and morphology. Although investigators differ in their findings concerning the relationship between age and gender and the corneal endothelial characteristics, the literature clearly shows a significant difference in corneal endothelial properties among races and ethnic groups [6 - 9].

In this study, the MCD of healthy senior Hispanic patients was found to be within normal values for healthy corneas [24]. Our mean number of cells studied per patient is higher than reported by other authors.

The MCD among various elderly populations from other countries is shown in Table **3**. Although none of the authors studied exclusively elderly patients, a stratified calculation based on their age groups was performed. The MCD of 2198.92 cells/mm^2 found in our study across all age groups is similar to the Caucasian population in Lithuania (2366 cells/mm^2) [1], the Turkish population studied by Goktas *et al*. [10] (2215 cells/mm^2), and distant from the findings of Hashemian *et al*. [11] in Iran (1681.61 cells/mm^2), and also from Padilla *et al*. in the Philippines (2780.45cells/mm^2) [6].

It is important to mention the results of Graue *et al*. [12] and Molina *et al*. [13] in the Mexican population from Central Mexico, where patients in the 60-89 years age group showed an MCD of 1805.33 cells/mm^2 and 1910.42 cells/mm^2, respectively, contrasting with our findings and suggesting that corneal endothelial cells characteristics may differ with our counterparts in the central and southern region of the country. Another study in a population from Central Mexico showed an MCD of 1909 cells/mm^2 in patients with a mean age of 71 years; interestingly, this cohort of patients had unilateral pseudophakic bullous keratopathy that developed after cataract surgery [25]. These geographical differences in MCD among the Mexican population may be clinically significant. However, further studies are needed to fully understand the mechanisms behind this disparity.

No decrease in MCD was found in our population as patients got older, suggesting that Hispanic patients who

maintain ocular health after 65 years can also preserve a fair number of endothelial cells. The latter is supported by the fact that 73% of patients had a cell density of over 2000 cells/mm^2, and only 12.45% showed an MCD <1500 cells/mm^2. It has been shown in previous reports that as the mean population's age studied increases, there is also an increased spread in the range of MCD counts, making the endothelial cell density measurement an unreliable index for the evaluation of corneal aging. The same finding applies to other animal species studied, including dogs, cats, monkeys, and rabbits, where the density and morphology of endothelial cells change with age, but the adult MCD remains constant [22, 23, 26].

As for the endothelial cell size and shape, our population showed a high degree of polymegethism and pleomorphism (MCV of 42.89% and PHC 46.25%). These findings correlate with the fact that in most populations studied, pleomorphism and polymegethism tend to increase as patients age [27 - 30] and suggest that elderly Hispanic patients could have higher rates of pleomorphism and polymegethism than other ethnicities [1, 6 - 13]. Surprisingly, the group of ≥85 years showed a fair rate of polymegethism and pleomorphism (51.85% and 44.4%, respectively).

Regarding CCT 527.55 (95% CI 524 to 531), we find it interesting that these values are similar to those reported in a previous study performed by our group in younger patients (mean age: 32.54 ± 12.04 years and Mean CCT: 545.69 ± 36.88 μm). The findings reported in both studies support the idea that the Mexican population could present a different mean CCT, and "normal" CCT could be redefined for clinical purposes in our context [31].

Further, 17% of eyes had cornea guttata. An exploratory analysis of these eyes revealed an MCD of 1611.63 cells/mm^2 and a mean CCT of 537.75, suggesting that cornea guttata was only a finding in these asymptomatic patients, and guttae might be associated to lower MCD values.

Some authors have found differences between gender in endothelial cell parameters [6 - 9]. As resumed in Table **2**, our results show that those differences are only statistically significant for MCD.

Limitations of this study are the small but otherwise healthy senior population, and the study's retrospective nature. Another further consideration is the possible presence of selection bias since specular microscopy is not performed as a routine examination in our country, and the cost may be prohibitively expensive to some patients.

CONCLUSION

However, we believe further prospective studies with control groups may be beneficial within the same population. This may be eased as specular microscopy becomes more available for the Mexican population.

In conclusion, the endothelial cell characteristics of elderly Mexican Northeastern patients differ from their counterparts from central and south Mexico and other nations. A high prevalence of polymegethism, pleomorphism, and guttata is present, but not in the >85-year age group, where a lower rate of these characteristics was described within the sample size studied.

AUTHORS' CONTRIBUTIONS

All authors fulfilled ICJME criteria of authorship.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study was approved by the Ethics and Research Committees (#ARCC/ECH2019).

HUMAN AND ANIMAL RIGHTS

The study was conducted following the Declaration of Helsinki principles.

CONSENT FOR PUBLICATION

All patients enrolled in the study participated voluntarily after reading and signing a previously written informed consent.

STANDARD OF REPORTING

STROBE guidelines have been followed in this study.

AVAILABILITY OF DATA AND MATERIALS

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

FUNDING

This research project received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

CONFLICT OF INTEREST

The authors declare no competing interests.

ACKNOWLEDGEMENTS

None.

REFERENCES

[1] Galgauskas S, Norvydaitė D, Krasauskaitė D, Stech S, Ašoklis RS. Age-related changes in corneal thickness and endothelial characteristics. Clin Interv Aging 2013; 8: 1445-50. [http://dx.doi.org/10.2147/CIA.S51693] [PMID: 24187493]

[2] Duman R, Tok Çevik M, Görkem Çevik S, Duman R, Perente İ. Corneal endothelial cell density in healthy Caucasian population. Saudi J Ophthalmol 2016; 30(4): 236-9. [http://dx.doi.org/10.1016/j.sjopt.2016.10.003] [PMID: 28003782]

[3] Krachmer JH, Mannis MJ, Holland EJ. Cornea. Elsevier Mosby 2011.

[4] Velazquez JS, Martinez FC, Brando VAC, *et al.* Automatic image processing applied to corneal endothelium cell count and shape characterization. Dyna (Bilbao) 2020; 95(1): 170-4. [http://dx.doi.org/10.6036/9275]

[5] Nelson LR, Hodge DO, Bourne WM. Central corneal endothelial cell changes over a ten-year period. Am J Ophthalmol 1997; 124(2): 273-4. [http://dx.doi.org/10.1016/S0002-9394(14)70810-4]

[6] Padilla MDB, Sibayan SAB, Gonzales CSA. Corneal endothelial cell density and morphology in normal Filipino eyes. Cornea 2004; 23(2): 129-35. [http://dx.doi.org/10.1097/00003226-200403000-00005] [PMID: 15075881]

[7] Snellingen T, Rao GN, Shrestha JK, Huq F, Cheng H. Quantitative and morphological characteristics of the human corneal endothelium in relation to age, gender, and ethnicity in cataract populations of South Asia. Cornea 2001; 20(1): 55-8.

[http://dx.doi.org/10.1097/00003226-200101000-00011] [PMID: 11189005]

[8] Matsuda M, Yee RW, Edelhauser HF. Comparison of the corneal endothelium in an American and a Japanese population. Arch Ophthalmol 1985; 103(1): 68-70. [http://dx.doi.org/10.1001/archopht.1985.01050010072023] [PMID: 3977679]

[9] Rao SK, Ranjan Sen P, Fogla R, Gangadharan S, Padmanabhan P, Badrinath SS. Corneal endothelial cell density and morphology in normal Indian eyes. Cornea 2000; 19(6): 820-3. [http://dx.doi.org/10.1097/00003226-200011000-00012] [PMID: 11095057]

[10] Goktas A, Gumus K, Mirza GE, Crockett C, Karakucuk S, Cavanagh HD. Corneal endothelial characteristics and central corneal thickness in a population of Turkish cataract patients. Eye Contact Lens 2012; 38(3): 142-5. [http://dx.doi.org/10.1097/ICL.0b013e318243e7d2] [PMID: 22223161]

[11] Hashemian MN, Moghimi S, Fard MA, Fallah MR, Mansouri MR. Corneal endothelial cell density and morphology in normal Iranian eyes. BMC Ophthalmol 2006; 6(1): 9. [http://dx.doi.org/10.1186/1471-2415-6-9] [PMID: 16519812]

[12] Graue-Weichers L, Valdez-Garcıa J, Ramırez-Luquın T, Claros A. Densidad celular endotelial. Estudio en población general de la Ciudad de México. Revista Mexicana de Oftalmología 1989; 63: 91-5.

[13] Rey DFM, Gómez A. Evaluation by decades of age behavior of corneal endothelial cells in population Mexican. Revista Mexicana de Oftalmología 2005; 792: 93-100.

[14] Geroski DH, Matsuda M, Yee RW, Edelhauser HF. Pump function of the human corneal endothelium. Effects of age and cornea guttata. Ophthalmology 1985; 92(6): 759-63. [http://dx.doi.org/10.1016/S0161-6420(85)33973-8] [PMID: 2412197]

[15] Bahn CF, Glassman RM, MacCallum DK, *et al.* Postnatal development of corneal endothelium. Invest Ophthalmol Vis Sci 1986; 27(1): 44-51. [PMID: 3941037]

[16] Laule A, Cable MK, Hoffman CE, Hanna C. Endothelial cell population changes of human cornea during life. Arch Ophthalmol 1978; 96(11): 2031-5. [http://dx.doi.org/10.1001/archopht.1978.03910060419003] [PMID: 718491]

[17] Ayala G, Díaz ME, Martínez-Costa L. Granulometric moments and corneal endothelium status. Pattern Recognit 2001; 34(6): 1219-27. [http://dx.doi.org/10.1016/S0031-3203(00)00074-1]

[18] Joyce NC, Harris DL, Mello DM. Mechanisms of mitotic inhibition in corneal endothelium: contact inhibition and TGF-beta2. Invest Ophthalmol Vis Sci 2002; 43(7): 2152-9. [PMID: 12091410]

[19] Smith JR, Pereira-Smith OM. Replicative senescence: implications for in vivo aging and tumor suppression. Science 1996; 273(5271): 63-7. [http://dx.doi.org/10.1126/science.273.5271.63] [PMID: 8658197]

[20] Norsgaard H, Clark BF, Rattan SI. Distinction between differentiation and senescence and the absence of increased apoptosis in human keratinocytes undergoing cellular aging in vitro. Exp Gerontol 1996; 31(5): 563-70. [http://dx.doi.org/10.1016/0531-5565(96)00011-3] [PMID: 9415111]

[21] Blake DA, Yu H, Young DL, Caldwell DR. Matrix stimulates the proliferation of human corneal endothelial cells in culture. Invest Ophthalmol Vis Sci 1997; 38(6): 1119-29. [PMID: 9152231]

[22] Faragher RGA, Mulholland B, Tuft SJ, Sandeman S, Khaw PT. Aging and the cornea. Br J Ophthalmol 1997; 81(10): 814-7. [http://dx.doi.org/10.1136/bjo.81.10.814] [PMID: 9486017]

[23] Laing RA, Sanstrom MM, Berrospi AR, Leibowitz HM. Changes in the corneal endothelium as a function of age. Exp Eye Res 1976; 22(6): 587-94. [http://dx.doi.org/10.1016/0014-4835(76)90003-8] [PMID: 776638]

[24] Hoffer KJ, Kraff MC. Normal endothelial cell count range. Ophthalmology 1980; 87(9): 861-6. [http://dx.doi.org/10.1016/S0161-6420(80)35149-X] [PMID: 7413150]

[25] Valdez-Garcia J, Pauli K, Madrid-Valero G, Grawe-Weichers E. Automated morphometric analysis of the contralateral eye in pseudophakic bullous keratopathy. Rev Mex Oftalmol 2000; 74: 267-70.

[26] Valdez-Garcia JE, Lozano-Ramirez JF, Zavala J. Adult white New Zealand rabbit as suitable model for corneal endothelial engineering. BMC Res Notes 2015; 8(1): 28. [http://dx.doi.org/10.1186/s13104-015-0995-1] [PMID: 25648773]

[27] Sawa M. The morphometry of the human corneal endothelium and follow-up of postoperative changes. Jpn J Ophthalmol 1979; 23(3): 337-50.

[28] Suda T. Mosaic pattern changes in human corneal endothelium with age. Jpn J Ophthalmol 1984; 28(4): 331-8. [PMID: 6530835]

[29] Yee RW, Matsuda M, Schultz RO, Edelhauser HF. Changes in the normal corneal endothelial cellular pattern as a function of age. Curr Eye Res 1985; 4(6): 671-8. [http://dx.doi.org/10.3109/02713688509017661] [PMID: 4028790]

[30] Carlson KH, Bourne WM, McLaren JW, Brubaker RF. Variations in human corneal endothelial cell morphology and permeability to fluorescein with age. Exp Eye Res 1988; 47(1): 27-41. [http://dx.doi.org/10.1016/0014-4835(88)90021-8] [PMID: 3409985]

[31] Valdez-García JE, Hernandez-Camarena JC, Lozano-Ramírez JF, Zavala J, Loya-García D, Merayo-Lloves J. Correlation of age, corneal curvature and spherical equivalent with central corneal thickness. Revista Mexicana de Oftalmología 2017; 91(4): 172-6. [http://dx.doi.org/10.1016/j.mexoft.2016.05.005]

© 2022 Valdez-García *et al*.

This is an open access article distributed under the terms of the Creative Commons Attribution 4.0 International Public License (CC-BY 4.0), a copy of which is available at: https://creativecommons.org/licenses/by/4.0/legalcode. This license permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.