Corneal Endothelial Cell Changes in Type 2 Diabetic Patients

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ABSTRACT

Background: Diabetic eye disease is an end-organ response to the effects of the condition on the human system. The cornea is a transparent structure protecting the anterior one-sixth of the eye. Chronic hyperglycemia can affect the corneal layers shape and functions that responsible for corneal transparency.

Objectives: Comparison of the changes in corneal endothelium namely, cell density (CD), percentage polymegathism, cell volume (CV) and pleomorphism (6A) along with central corneal thickness (CCT) in patients of type 2 diabetes mellitus with normal age and sex- matched subjects.

Patients and methods: A case-control study in Sayed Galal University Hospital. 20 eyes of type 2 diabetic patients and 20 eyes of age and sex matched controls were included in the study. Age, gender, other demographic data, and relevant diabetic history were obtained. Full evaluation of both cases and controls including both anterior and posterior segment complete evaluation was done. Non- contact specular microscopy was used to study the corneal endothelial cells.

Results: The mean endothelial cell density (ECD) was non-significantly lower in type 2 diabetics compared to controls (p value = 0.751.). There was a highly significant reduction in hexagonally in the cases compared to controls and highly significant increase in coefficient of variation in the cases compared to controls

Conclusion: This study acknowledged that type 2 diabetes causes a significant decrease of hexagonality and increase CV (polymegathism). However, non-significant reduction of ECD was documented.

Keywords: Diabetes; Cornea; Endothelial cell changes; Specular Microscopy.

INTRODUCTION

Diabetes is one of the major non-infective diseases in the globe in the current millennium. The International Diabetes Federation recorded its prevalence to be 246 million in 2007 and may reach to 380 million by 2025 [1]. Type II Diabetes mellitus is a metabolic ailment characterized by elevated blood glucose due to insulin resistance accompanied by relative insulin deficiency. Diabetes mellitus types II represent 90% of diabetics while 10% primarily due to type I and gestational diabetes. Diabetic eye disease is an end-organ response to the effects of the condition on the human systems. The cornea is a transparent structure protecting the anterior one-sixth of the eye [2].

Chronic hyperglycemia can affect the corneal layers shape and functions that responsible for corneal transparency [3]. Corneal endothelium is the innermost layer of hexagonal non-replicating neural crest-derived tissue that is responsible for maintaining corneal transparent throughout life by pumping excess fluid out of the stroma and keeping stroma in its usual dehydrated state [4]. Corneal endothelial cell count is vital for maintaining corneal clarity [5].

Non-contact Specular microscopy is a non-invasive method of morphological analysis of the corneal endothelium. It enables the measurement of mean cell count. This provides an index of the corneal endothelial layer function.

PATIENTS AND METHODS

This study was planned to be case-control study assessing forty eyes. Comparing mean corneal endothelial cell count changes between type II diabetic patients, and healthy adults. They were subdivided into two groups:

1) 20 eyes (cases) of diabetic patients.
2) 20 eyes (control) of healthy adults.

Each case had a full ocular examination including uncorrected visual acuity and BCVA then the intraocular pressure was determined by Goldmann Applanation Tonometry. Anterior segment evaluation was done by use of the slit lamp and posterior segment evaluation by use of the ninety diopters auxillary lens.

Slit lamp examination was done to rule out any local disease that affect the cornea.

Specular microscopy of corneal endothelium was carried out using the noncontact Topcon model SP-1P non-contact specular microscope.

These machines can auto-tract the cornea and autofocus on the endothelium without touching the cornea. It also provides high magnification, good quality image. It has semi-automated, computer-assisted cell count assessment and morphometric evaluation.

Taking three images; central, nasal and temporal. The "panorama" function of the SP-1P automatically combines those images creating a large area for the observation and analysis of endothelial cells, and then analyze the cell sizes according to various factors that includes cell density, the CV of cell area (SD/mean), hexagonality. About 100 cells were counted in each image analysis.

Further morphometric analysis and automated cell analysis was done to obtain mean corneal endothelial analysis and automated cell analysis was done to obtain mean corneal endothelial cell density (ECD,

cells/mm²), coefficient of variation (CV), hexagonality (6A), central corneal thickness (CCT).

All the enrolled cases were examined by the same ophthalmogist.

The study was approved by the Ethics Board of Al-Azhar University and an informed written consent was taken from each participant in the study.

RESULTS

Demographics of control and diabetic patients (eyes) are shown in Table (1).

- **In non-diabetic group**, the mean age was 56.55 ± 5.35 years old ranged from 45-63 years old. 55% were females and 45% were males. 50% of enrolled eyes were right and 50% of them were left.
- **In diabetic group**, the mean age was 56.45 ± 6.22 years old ranged from 43-64 years old. 70% were males and 30% were females. 45% of enrolled eyes were right and 55% of them were left.

Table (1): Demographic data of both groups.

		Non diabetic group	Diabetic group	Test value	P-value	Sig.
		No. = 20	No. = 20			
Age	Mean ± SD	56.55 ± 5.35	56.45 ± 6.22	-0.055•	0.957	NS
	Range	45 – 63	43 – 64			
Sex	Female	9 (45%)	6 (30%)	0.960*	0.327	NS
	Male	11 (55%)	14 (70%)			
Site	OD	10 (50%)	9 (45%)	0.100*	0.752	NS
	OS	10 (50%)	11 (55%)			

Endothelial cell changes included changes in CD, CV and HEX. Mean endothelial cell count in control group and diabetic group was 2868.1 ± 295.43 and 2831.65 ± 415.5 respectively as shown in table (2). Independent samples t-test was used to compare mean endothelial cell count which showed non-significant statistical decrease in diabetic group than control group as p-value of CD was 0.751 (P > 0.05).

The CV was 32.8 ± 3.04 in non-diabetic group and 37.7 ± 1.98 in diabetic group as shown in table (2). Independent samples t-test was used to compare CV, which showed highly significant statistical increase in diabetic group as p-value of CV was 0.000 (P<0.001), which means that CV in type 2 diabetic patients was more compared to healthy adults.

The HEX was 38.05 ± 4.63 in non-diabetic group and 30.9 ± 6.48 in diabetic group as shown in table (2). Independent samples t-test was used to compare HEX, which showed highly significant statistical decrease in HEX in diabetic group as p-value of HEX was 0.000 (P < .001), which means that HEX in type 2 diabetic patients was less compared to healthy adults (Table2)

Table (2): Comparison of corneal endothelial cell changes among cases and control.

		Non diabetic group	Diabetic group	Test value•	P-value	Sig.
		No. = 20	No. = 20			
CD	Mean ± SD	2868.1 ± 295.43	2831.65 ± 415.51	-0.32	0.751	NS
	Range	2249 – 3304	2371 – 3887			
CV	Mean ± SD	32.8 ± 3.04	37.7 ± 1.98	6.048	0	HS
	Range	29 – 38	34 – 43			
HEX	Mean ± SD	38.05 ± 4.63	30.9 ± 6.48	-4.016	0	HS
	Range	31 – 47	15 – 44			

DISCUSSION

The corneal posterior surface is covered by a single layer of polygonal cells known as corneal endothelium. It has the lowest mitotic activity. The main function of the endothelium is providing regulation of dehydration state of the cornea through ATP and bicarbonate-dependent pump and thereby maintenance of its detergence. In the adult human, the normal cell density is 2600 -3000 cells/mm^2 and the percentage of hexagonality is 60 - 75%. The central cell density diminishes by increasing age at an average rate of 0.6% every year. To maintain its proper structure and function, endothelial cells shows stretching and migration to cover injured area in response to minor damage, which in turn lead to polymegathism and pleomorphism [6].

Diabetes mellitus causes morphological changes in the corneal endothelial cell. This is known as pleomorphism and polymegathism [7]. According to our results ECD, CV, HEX in type 2 diabetic patients were not quite the same as control group and we noticed a few relations with some of our measured parameters.

In this study, the mean ECD was found to be not significantly lower in patients with type 2 diabetes mellitus when compared to controls. (p value = 0.751). This was also noted by **Itoi *et al.*** [7] **Matsuda *et al.*** [8] **and Storr-Paulsen *et al.*** [9] where they did not find any statistically significant difference with regard to endothelial cell density. But studies by **Sumit *et al.*** [10] involving more sample size (540 or less) and **Rizvi and Zafar** [11] involving more sample size (130), they found that the mean CD was significantly lower in patients with type 2 diabetes mellitus relative to controls. Also, **El-Agamy and Alsubaie** [12]**, Sudhir *et al.*** [13], **Choo *et al.*** [14] **and Inoue *et al.*** [15] studies acknowledged a significant decrease in ECD of diabetic corneas compared to controls. But, a study by **Siribunkum *et al.*** [16] showed significantly more increased corneal endothelial cell density in diabetic patients.

This study assessed coefficient of variation and hexagonality in both groups. It showed a highly significant polymegathism in diabetic cornea. This is similar to studies by **El-Agamy and Alsubaie[12]**, **Shenoy et al. [17] and Lee et al. [18]**.This increase indicated that endothelial cells enlarged to fill the gaps between neighboring cells. In opposition, **Sumit et al. [10]**, **Inoue et al. [15] Chen et al. [19] and Sudhir et al. [20]** reported that they did not detect any significant polymegathism in diabetic compared to non-diabetic corneas. Many studies **Choo et al. [14] and Sudhir et al. [20]** explained the morphological characters of diabetic corneas. This was done through assessment of polyol (sorbitol–aldose reductase) pathway in diabetic corneas. They showed that high levels of glucose lead to expanded action of the aldose reductase that caused sorbitol buildup in both epithelial and endothelial cells of the cornea. Sorbitol acts as an osmotic agent causing augmentation of endothelial cells. Furthermore, diabetes reduces Na+–K+ ATPase activity of the corneal endothelium, and lead to changes in the morphology and permeability in diabetic cornea, resulting in corneal damage. In addition, pump function of the endothelium is affected by ATP production reduction resulting from slowing down the Krebs cycle in cornea of diabetic patient.

This study showed a highly significant polymorphism in diabetic cornea compared to controls. This also was founded by **Choo et al. [14] and Lee et al. [18]** studies. In opposition**, El-Agamy and Alsubaie [12]**, **Storr-Paulsen et al.[9], Sudhir et al. [13] and Inoue et al. [15]** studies that verified non-significant difference between both groups.

CONCLUSION

Specular microscopes are used to estimate the structure and function of corneal endothelial cells. In this study, a highly significant reduction in percentage of hexagonality in the cases compared to controls. Besides, there was a highly significant increase in coefficient of variation in the cases compared to controls. Non-significant reduction in endothelial cell density in cases was found but considerable rise of central corneal thickness among diabetic subjects compared to healthy adults was observed. The study further suggests conducting more studies, so that if more studies proven the changes, then this evaluation should become a part of the protocol for eye care in diabetic population in Egypt.

RECOMMENDATIONS

In further studies, correlation of severity of diabetic retinopathy in relation to endothelial cell changes and central corneal thickness can be assessed and changes in diabetic type 2 compared to type 1 can be done.

It is not clear whether there is a direct relationship between endothelial cell count between cases and controls. In **Rizvi and Zafar [11],** the mean endothelial cell count in type 2 diabetic patients was significantly less as compared to healthy adults. This might be because of large number of eye compared to our study.

In our study, patients who had laser photocoagulation were excluded because of its effect on the corneal structures. This resulted in a small sample size. Therefore, we recommend further studies with a larger sample size to verify the extent of corneal endothelial changes caused by type 2 DM.

Correlation of HbA1c level changes in diabetic subjects and corneal changes should be also studied.

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