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Observational Study

Tear inflammation related indexes after cataract surgery in elderly patients with type 2 diabetes mellitus

INTRODUCTION

Cataracts are the main cause of blindness and affect millions of people worldwide[1]. Diabetes is one of the most prevalent chronic diseases in the world. Patients with type 2 diabetes have a higher risk of cataracts than those without diabetes and require surgery more urgently[2]. Cataract patients with diabetes are also at a higher risk of delayed incisional healing and postoperative complications, such as dry eye, corneal epithelial defects or erosions, persistent inflammatory reactions, and infections[3]. Currently, China is rapidly becoming an aging society, with an increasing proportion of the aged population. Changes in tear composition in elderly patients resulting from loss of the meibomian gland gradually aggravate with age. Additionally, abnormal diabetes-induced variations in tear components might cause postoperative inflammatory reactions in patients with type 2 diabetes[4]. Xerophthalmia was observed significantly more frequently in diabetic patients than in non-diabetics 7 d after phacoemulsification[5]. Another retrospective clinical study confirmed that the risk of complications in patients with diabetes was highest in the first 2 wk after cataract surgery[6].

The development of postoperative inflammation may be significantly affected by these inflammation-related mediators, but quantitative studies on inflammatory-related content changes in tears, particularly the effect of diabetes mellitus, are still lacking. This study focused on the postoperative expression of inflammatory factors in elderly

diabetic cataracts to discuss the effects of drugs on the control of postoperative inflammation.

MATERIALS AND METHODS

Patients

Patients diagnosed with age-related cataracts and treated with cataract surgery in our hospital between December 2021 and January 2022 were divided into group A (cataract with combined type 2 diabetes mellitus, n = 20 eyes) and Group B (elderly patients with cataracts but no diabetes, n = 20 eyes).

The inclusion criteria were as follows: patients with cataract with or without a confirmed history of type 2 diabetes mellitus, eligibility for geriatric cataract surgery, clear state of consciousness, and ability to cooperate with relevant examinations.

Exclusion criteria: Patients with previous/current ocular/systemic inflammation, fever, immunological diseases, history of ocular surgery or trauma, intraoperative complications, or inability to cooperate with examinations.

General clinical parameters, such as age, sex, body temperature, height, and weight, and detailed medical history were acquired, measured, and recorded. Hemanalysis and measurement of indicators were performed for all patients, including blood glucose, triglycerides (TG), total cholesterol (TC), glycated hemoglobin (HbA1c), glycated albumin (GA), tear matrix metalloproteinase-2 (MMP-2), MMP-9, tissue inhibitor of metalloproteinase-1 (TIMP-1), TIMP-2, interleukin-6 (IL-6), and IL-20. All patients underwent ophthalmic observations and examinations, including visual acuity (before and postoperative, categorized as ≤ 0.3, 0.3-0.6, and ≥ 0.6), intraocular pressure, slit lamp examination, fundus state, cataract-related preoperative examinations, ocular-surface states, healing and inflammatory states of corneal incision.

Tears collection and measurement

Tears were collected before surgery and on days 1 and 3 and weeks 1, 2, 3, and 4 post-operation. Saline (150 μL) was dropped into the conjunctival sac of the surgical eye,

followed by oculogyration in four directions. The fluid in the conjunctival sac was extracted using a sterile syringe and stored in Eppendorf tubes at -80°C until measurement. The concentrations of MMP-2, MMP-9, TIMP-1, TIMP-2, IL-6, and IL-20 in the tear fluid were measured using enzyme-linked immunosorbent assay.

Surgical procedure

All patients were administered pranoprofen eye drops 3 times/d and levofloxacin eye drops 3 times/d, three days before surgery. All procedures were performed by the same surgeon. Mydriasis was induced with compound tropicamide 30 min before surgery and surface anesthesia with oxybuprocaine hydrochloride drops before surgery. A main incision was made on the temporal side of the transparent cornea, and a secondary incision was made on the inferior temporal (left eye) or superior temporal (right eye) side of the transparent cornea. Continuous circular capsulorhexis was performed through the injection of viscoelastic agents, the nucleus was emulsified after hydro-dissection and hydro-delineation, followed by aspiration of the cortex, and the intraocular lens was implanted into the polished capsular bag. Surgery was completed after irrigation of the anterior chamber, aspiration of viscoelastic agents, and closure of the conjunctiva with solution. After surgery, all patients were administered tobramycin and dexamethasone eye drops three times/d for one month, pranoprofen eye drops three times/d for two weeks, and levofloxacin eye drops three times/d for two weeks.

Statistical analysis

Differences in inflammatory factor expression (indicated as mean and standard deviation) between diabetic and non-diabetic elderly patients with cataract were determined by performing repeated-measures and Analysis of Variance using SPSS 26.0. Differences in age, intraocular pressure (IOP), HbA1c, GA, TG, and TC between the two groups were verified using Student's test in SPSS 26.0. Differences in sex between the two groups were determined using the χ^2 test. Statistical significance was set at $P < 0.05$.

RESULTS

Comparison of general information

A comparison was performed with 19 eyes of 19 males (47.5%) and 21 eyes of 21 females (52.5%), whose mean age was (70.3 ± 6.3) years, and the mean disease course duration of diabetes in group A was (6.8 ± 2.2) years. Patients were further grouped based on their preoperative visual acuity as ≤ 0.1, 0.1-0.3, and ≥ 0.3. The composition of sex and age, visual acuity, IOP, TG, and TC between the groups was not significantly different, while significant differences were detected in HbA1c and GA (Table 1).

Changes in the expression levels of MMP-2 and MMP-9 in tear fluid at each time point in the two groups

The level of MMP-2 in both groups continuously increased until it peaked in the first week postoperatively and then gradually decreased over the next three weeks, ultimately declining to a level lower than the preoperative level at week 4. The level of MMP-9 peaked in the first two weeks postoperative and then returned to the same level as 1-day post-operation. The expression levels of MMP-2 and MMP-9 in group A were significantly higher than those in group B at all time points (Table 2, Figure 1A and B; $P < 0.001$).

Changes in the expression levels of TIMP-1 and TIMP-2 in tear fluid at each time point in the two groups

After a decline in the first two postoperative weeks and an increase from the third week, the concentration of TIMP-1 in group A was still lower than that before surgery at four weeks post-operation. The expression level of TIMP-1 in group A was lower than that in group B (Figure 1C, $P < 0.05$). The level of tear TIMP-2 in group A was higher than that in group B before and after operation (Table 3, Figure 1D; $P < 0.01$).

Changes in IL-6 and IL-20 expression levels in tear fluid at each time point in both groups

After surgery, IL-6 Levels in both groups increased in the first week, but remained at a higher level in group A than in group B (Figure 1E, $P < 0.001$). Similar trends in IL-20 Levels were observed in the two groups, which were also higher in group A than in group B ($P < 0.05$). Its concentration remained constant before the third week after operation, surged to a peak in the third week post-operation, and then started to slump in the fourth week (Table 4, Figure 1F).

DISCUSSION

Hyperglycemia contributes to impaired corneal sensitivity, reduces nerve fiber density, and delays epithelial wound healing. Due to reduced corneal sensitivity, reflex-induced tear secretion decreases together with the blink rate in diabetic patients, which ultimately leads to increased tear evaporation[7]. Corneal incision accompanied by nerve amputation and microscopic light illumination in cataract surgeries, use of anesthetics, mydriatic drops, and postoperative antibiotics and hormones increases the risk of postoperative complications in diabetic patients. In summary, patients with type-2-diabetes with cataracts are at a higher risk of postoperative complications and have more difficulty in epithelial wound healing than cataracts in patients with normal blood glucose levels, which suggests that more attention should be paid to their treatment.

MMPs are a highly conserved family of proteinases that can degrade various extracellular matrix components[8]. The expression levels of MMPs are extremely low under normal physiological conditions and can be significantly upregulated by inflammatory factors, growth factors, and pathological conditions such as high glucose and oxidative stress. TIMPs are active in many tissues and body fluids as endogenous inhibitors of MMPs[9]. It was confirmed both *in vitro* and *in vivo* that upregulated expression levels of MMP-2 and MMP-9 in wound healing of high glucose cultured corneal epithelial cells and corneal epithelial cells from diabetic rats can lead to xerophthalmia, defects, and erosions of corneal epithelial and ocular inflammation[10].

Increased MMP-9 expression in ocular tissues has also been observed in recurrent corneal erosion, skin ulcers, and diabetic retinopathy[11]. Tears containing levels of MMP-2, MMP-9, and TIMP-2 before and post-operation, were estimated to be higher in patients with diabetes than in elderly patients with cataracts but no diabetes. It is thought to be a response to the stimulation of the ocular surface by long-term high blood glucose concentrations and chronic inflammation. In addition, the gradual increase in MMP-2/9 Levels in the first two postoperative weeks suggested that severe inflammatory responses occurred in the first two weeks post cataract surgery. TIMP-1 expression was suppressed after surgery in both groups and was more significant in group A. This suppression works in concert with the upregulated expression of MMPs and ultimately causes severe inflammation in patients with diabetes.

IL-6 is a pleiotropic cytokine that affects various cell types, including pro-inflammatory and anti-inflammatory cytokines[12]. Dysregulation of IL-6 signaling is associated with the pathogenesis of several autoimmune and inflammatory diseases, including type 2 diabetes[13]. The causality between chronic low-grade inflammation, indicated by elevated circulating levels of inflammatory cytokines (*e.g.*, IL-6), and the pathogenesis of type 2 diabetes has been progressively verified[14]. Previous studies have shown that during trauma, many inflammatory cells accumulate and release early inflammatory mediators, mainly tumor necrosis factors-α and IL-6, which initiate a systemic inflammatory response and promote the expression of MMP-2, the overexpression of which is responsible for the disease. In corneal keratopathy, IL-6-mediated MMP-2 expression results in continuous tissue necrosis followed by degradation[15].

The interaction between IL-20 and its receptor may have pro-inflammatory, angiogenic, and chemo-attractive effects in chronic inflammatory diseases, especially atherosclerosis and rheumatoid arthritis. This may also have a certain degree of impact on type 2 diabetes. We also detected the expression of IL-20 and related receptors in corneal epithelial cells, dendritic cells, and monocytes of wild-type mice. By promoting the aggregation and activation of T-cells in the injured cornea, IL-20 exerts anti-

inflammatory effects without increasing neutrophil chemotaxis or promoting corneal epithelialization and wound healing[16]. This process of corneal re-epithelialization can be inhibited by the absence of neutrophils or T cells. In this study, IL-6 Levels gradually increased to a peak on days 1 and 3; and on week 1 post-operation, and then gradually decreased at weeks 2, 3, and 4 post-operation. This might be related to the gradual aggravation of early inflammation, which could induce the expression of IL-6 to further promote anti-inflammatory effects after cataract surgery. The increase in IL-20 in the third week after cataract surgery might be caused by the decreased release of inflammatory factors in the third week after cataract surgery, which could promote IL-20 expression and further contribute to corneal wound healing.

In this study, as there was a trend of correlated changes in postoperative inflammatory factor expression when the same ophthalmic medication was applied pre and postoperatively to the eyes of both groups, it was speculated that the application of anti-inflammatory and infection-preventive ophthalmic drugs before and after surgery had an effect on postoperative healing. Meanwhile, both the pre and postoperative levels of relevant inflammatory factors were higher in the test group than in the control group, indicating that the postoperative inflammatory response was higher in the test group based on the application of the same dosages of ophthalmic drugs. Therefore, it was considered clinically that within one week after cataract surgery, the frequency and duration of relevant ophthalmic drugs could be increased to reduce the postoperative inflammatory response in patients with combined diabetes and cataracts. Another study found that the use of ultrasound emulsification combined with IOL implantation based on routine glycemic control, IOP control, and anti-inflammation in patients with cataracts combined with diabetes, reduced the levels of inflammatory factors in the atrial fluid and oxidative stress indicators in such patients[17].

Our study has several limitations. First, it was a small sample; second, there was a lack of information about the patients' blood glucose levels and the duration of their disease, and some patients may have been undiagnosed or were untreated for diabetes

before surgery; third, the number of preoperative tears and tear volume in patients was inadequate.

CONCLUSION

Comparison between inflammatory indices at different time points before and after surgery revealed more severe postoperative inflammation in patients with Type 2 diabetes with cataracts than in elderly patients with cataracts but without diabetes. Postoperative levels of inflammatory factors in tears were fluid, particularly compared to levels before-the operation. The expression of most inflammatory factors peaked in the first two weeks after surgery, when patients were considered most vulnerable to inflammatory complications. Therefore, the increased use of anti-inflammatory drugs in the first two postoperative weeks was proposed based on our observations.

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Figure Legends

Figure 1 Changes of some indicators' levels in tears before and after surgery in both groups. A: Matrix metalloproteinase-2 (MMP-2); B: MMP-9; C: Tissue inhibitor of metalloproteinase-1 (TIMP-1); D: TIMP-2; E: Interleukin-6 (IL-6); F: IL-20. $^{a}P < 0.05$, $^{b}P < 0.01$, $^{c}P < 0.001$. MMP: Matrix metalloproteinase; TIMP: Tissue inhibitor of metalloproteinase; IL: Interleukin.

Table 1 Comparison of general information between two groups of patients

Groups	Age[1] (yr)	Gender[2] (M/F)	Visual acuity (BCVA)[3]			Intraocular pressure[1] (mmHg)	HbA1c[1] (%)	GA[1] (%)	TG[1] (mmol/L)	TC[1] (mmol/L)
			≤ 0.1	0.1-0.3	≥ 0.3					
Group A	69.3 ± 6.6	9/11	9	8	3	15.3 ± 2.28	8.2 ± 0.6	25.1 ± 4.8	2.0 ± 0.3	5.7 ± 0.4
Group B	71.0 ± 5.0	10/10	8	10	2	15.8 ± 2.76	5.4 ± 0.1	14.0 ± 1.5	1.7 ± 0.3	5.6 ± 0.4
χ^2/F value	1.196	0.100		0.481		0.225	8.197	8.700	0.238	0.749
P value	0.557	0.752		0.829		0.575	0.002	0.020	0.458	0.142

[1]The use of two independent samples *t*-test.

[2]The use of the χ^2 test.

[3]The use of Pearson's χ^2 test.

HbA1c: glycated hemoglobin; GA: glycated albumin; TG: triglycerides; TC: total cholesterol.

Table 2 Comparison of matrix metalloproteinase-2 and matrix metalloproteinase-9 levels in the tears of two groups at different time points

Time	MMP-2 (ng/mL)				MMP-9 (ng/mL)			
	Group A	Group B	*t* value	*P* value	Group A	Group B	*t* value	*P* value
Preoperative	11.13 ± 0.56	8.83 ± 0.88	11.65	0.000	36.07 ± 1.82	25.55 ± 1.74	13.22	0.000
1 d	10.71 ± 0.68	8.07 ± 0.68	10.54	0.000	42.90 ± 1.82	32.69 ± 2.33	10.96	0.000
3 d	13.53 ± 0.79	10.42 ± 0.96	11.06	0.000	43.37 ± 1.33	32.80 ± 1.02	18.09	0.000

1 wk	14.45 ± 0.9	10.54 ± 0.94	8.22	0.000	56.25 ± 1.96	43.02 ± 1.45	20.45	0.000
2 wk	13.17 ± 0.93	9.43 ± 0.49	12.29	0.000	72.78 ± 1.66	51.99 ± 1.71	41.48	0.000
3 wk	11.37 ± 0.40	9.15 ± 0.60	9.99	0.000	43.81 ± 2.68	32.55 ± 1.3	14.70	0.000
4 wk	8.77 ± 0.83	7.62 ± 0.84	2.63	0.017	44.41 ± 3.15	31.97 ± 1.58	13.79	0.000

MMP: Matrix metalloproteinase.

Table 3 Comparison of tissue inhibitor of metalloproteinase-1 and tissue inhibitor of metalloproteinase-2 levels in the tears of two groups of patients at different time points

Time	TIMP-1 (ng/mL)				TIMP-2 (ng/mL)			
	Group A	Group B	*t* value	*P* value	Group A	Group B	*t* value	*P* value
Preoperative	5.24 ± 0.13	5.77 ± 0.10	2.34	0.032	4.28 ± 0.15	3.33 ± 0.28	6.13	0.004
1 d	5.25 ± 0.15	5.76 ± 0.12	2.06	0.028	4.22 ± 0.18	3.58 ± 0.34	6.01	0.003
3 d	5.23 ± 0.14	4.89 ± 0.11	2.83	0.027	4.19 ± 0.13	3.71 ± 0.2	5.41	0.007
1 wk	4.57 ± 0.15	4.61 ± 0.23	0.45	0.060	4.28 ± 0.13	3.44 ± 0.36	5.08	0.006
2 wk	4.20 ± 0.13	5.51 ± 0.15	2.75	0.021	4.23 ± 0.18	3.51 ± 0.31	6.51	0.002
3 wk	4.71 ± 0.18	5.50 ± 0.14	5.75	0.005	4.29 ± 0.16	3.50 ± 0.35	6.60	0.004

4 wk	4.70 ± 0.17		5.77 ± 0.13		7.34	0.003	4.19 ± 0.16		3.50 ± 0.37		6.64	0.003

TIMP: Tissue inhibitor of metalloproteinase.

Table 4 Comparison of interleukin-6 and interleukin-20 levels in the tears of two groups of patients at different time points

Time	IL-6 (pg/mL)				IL-20 (pg/mL)			
	Group A	Group B	*t* value	*P* value	Group A	Group B	*t* value	*P* value
Preoperative	42.84 ± 1.49	29.89 ± 1.09	22.33	0.000	579.90 ± 13.89	533.15 ± 10.9	2.78	0.021
1 d	42.77 ± 1.18	30.17 ± 1.11	24.65	0.000	576.82 ± 10.67	535.13 ± 16.38	2.39	0.024
3 d	48.11 ± 2.01	33.79 ± 1.08	19.90	0.000	587.52 ± 7.62	534.28 ± 17.92	2.91	0.037
1 wk	53.85 ± 1.24	42.97 ± 0.52	25.76	0.000	578.75 ± 13.9	539.97 ± 11.95	2.50	0.038
2 wk	49.58 ± 2.02	37.82 ± 1.55	14.63	0.000	579.08 ± 11.15	534.64 ± 13.27	2.67	0.035
3 wk	42.64 ± 1.43	31.44 ± 1.57	16.73	0.000	702.67 ± 35.3	679.85 ± 26.2	2.62	0.032
4 wk	35.82 ± 1.14	29.79 ± 0.81	13.70	0.000	619.55 ± 60.04	570.05 ± 15.94	2.43	0.036

IL: Interleukin.

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