Expression of PCNA, ICAM-1, and Vimentin in Lens Epithelial Cells of Cataract Patients with and without Type 2 Diabetes

Haiyan FAN, Takahiro SUZUKI, Masafumi OGATA, Yoshihiro NAKAGAWA, Hiromi ONOUCHI and Kenji KAWAI

Department of Ophthalmology, Tokai University School of Medicine

(Received September 26, 2011; Accepted May 17, 2012)

Objective: In order to explore the pathological feature in the progression of cataract with type 2 diabetes, we compared the expression of proliferating cell nuclear antigen (PCNA), intercellular adhesion molecule-1 (ICAM-1), and vimentin in lens epithelial cells (LECs) of cataract patients with and without type 2 diabetes.
Methods: The indirect immunoperoxidase technique was performed on anterior capsules obtained from 25 patients with type 2 diabetes (DM group) and 25 patients without diabetes (control group). Immunohistochemical difference in the expression of PCNA, ICAM-1, and vimentin in LECs between the 2 groups was investigated.
Results: PCNA expression was decreased ($P = 0.036$) but ICAM-1 expression was significantly increased ($P < 0.0001$) in the DM group as compared with that in the control group. No difference was noted in the expression of vimentin between the 2 groups ($P = 0.264$).
Conclusions: Decreased proliferation of LECs and increased expression of ICAM-1 in LECs might play an important role in the progression of cataract with type 2 diabetes.

Key words: diabetic cataract, Intercellular adhesion molecule-1, lens epithelial cells, proliferating cell nuclear antigen, vimentin

INTRODUCTION

Diabetes, also referred to as diabetes mellitus (DM), is associated with a 5-fold higher prevalence of cataract, one of major causes of blindness in the world [1]. The adult-onset diabetic cataracts are readily formed in subjects with type 2 diabetes. It is well known that adult-onset diabetic cataracts may present with single cortex opacity, nuclear opacity, subcapsular opacity, or more than 2 kinds of opacities concurrently, which closely resemble age-related cataracts without diabetes. However, the nuclear and cortex hardness is different between adult-onset diabetic cataract and age-related cataract without diabetes even if the same opacities in nuclear or cortex are observed clinically. It suggests that adult-onset diabetic cataracts have more inherent alterations than age-related cataracts without diabetes in the progression of cataract.

Proliferating cell nuclear antigen (PCNA) is present in the late G1 and early S phases but not in the G0 phase of the cell cycle. PCNA expression level is a sensitive index of DNA synthesis and cell proliferation. In vivo and in vitro experiments have suggested that the decreased lens epithelial cell density observed in diabetes patients results from attenuated cell growth and induction of apoptosis [2–4].

Intercellular adhesion molecule-1 (ICAM-1), also known as CD54, is a 75–115-kDa cell surface glycoprotein. It is a member of the immunoglobulin super family of cell adhesion molecules, which preferentially mediate the attachment and transendothelial migration of circulating leukocytes, including monocytes, lymphocytes, and polymorphonuclear cells [5, 6]. ICAM-1 has been detected in LECs of cataract samples and is involved in the attachment and growth of LECs on collagen- or laminin-coated plates [7]. Recently, experimental studies have shown that fructose can directly increase the expression of ICAM-1 [8, 9].

Vimentin has been regarded as a marker of epithelial-mesenchymal transition (EMT). EMT is a central mechanism for diversifying the cells found in complex tissues. This dynamic process helps organize the formation of the body plan, and while EMT is well studied in the context of embryonic development, it also plays a role in the genesis of fibroblasts during organ fibrosis in adult tissues [10]. According to previous research, vimentin is low expressed in normal LECs; both vimentin and cytokeratin are over expressed in the LECs of nuclear cataract patients but only vimentin is over expressed in the LECs of anterior subcapsular cataract patients; the expression level of vimentin is higher in LECs of subcapsular cataract patients than that in LECs of nuclear cataract patients [11].

In our study, we hypothesized that the progression of cataract in patients with type 2 diabetes is because of abnormal proliferation, adhesion, migration, and transdifferentiation of LECs. Thus, we investigated the expression of PCNA, ICAM-1, and vimentin in the LECs of cataract patients with and without type 2 diabetes.

MATERIALS AND METHODS

Patients

Circular anterior capsules with attached LECs were

Haiyan FAN, Department of Ophthalmology, Tokai University School of Medicine, 143 Simokasuya, Isehara, Kanagawa 259-1193, Japan
TEL: +81-463-93-1121 FAX: +81-463-91-9328 E-mail: cn6283annie@yahoo.co.jp

obtained from 25 patients with type 2 diabetes (DM group) and 25 patients without diabetes (control group). The DM group was defined by the adult-onset cataract patients with type 2 diabetes. The control group was defined by the age-related cataract patients without diabetes. All the patients underwent cataract surgery at Tokai University Hospital from May 2010 to November 2010. Patients who were under steroidal antiinflammatory drug treatment and those who had systemic disorders such as carcinoma, rheumatic disease, and other chronic inflammatory diseases were excluded from this study. Patients who had ocular complications, such as pseudoexfoliation syndrome, high myopia of over -6.00 diopters (D), glaucoma, uveitis, trauma, and other retinal diseases, except diabetic retinopathy (DR), were also excluded from this study. None of the patients had undergone previous ocular surgery. All the clinical parameters were obtained just before the operation. The severity of different types of cataracts (cortical, nuclear, and posterior subcapsular cataract) was estimated according to The Lens Opacities Classification System Ⅲ (LOCS Ⅲ) [12]. The diagnosis of type 2 diabetes was made by a diabetologist at Tokai University according to the criteria of the American Diabetes Association (ADA) [13]. Informed consent was obtained from all patients enrolled in the study. The research followed the tenets of the Declaration of Helsinki. The study protocol was approved by the Clinical Trials Review Committee of Tokai University School of Medicine in 2010, before study activation.

Biopsy collection

One capsule was randomly selected if the patients underwent surgery of both eyes. The circular anterior capsules, approximately 5−6 mm in diameter, with attached LECs were obtained by performing continuous curvilinear capsulorhexis (CCC) with capsulorhexis forceps (Kawai Capsulorhexis Forceps; Asico, USA).

Immunohistochemistrystaining

Paraffin-embedded anterior capsular tissues fixed with 4% paraformaldehyde (PFA; pH 7.4) were sliced into sections of 3 μm thickness, and 100 sections were obtained for each subject. The method used for immunohistochemistry (IHC) staining was as follows. After deparaffinization and rehydration, heat-mediated antigen retrieval was done with citrate buffer (0.01 M, pH 6.0) in a microwave oven (98ºC, 10 min). The sections were incubated with 3% hydrogen peroxide for 10 min to quench endogenous peroxidase activity and then blocked with 10% normal goat serum for 20 min at room temperature (RT). The sections were incubated with mouse monoclonal anti-human PCNA (dilution 1:400; Dako, Japan) for 60 min at RT or mouse monoclonal anti-human ICAM-1 (dilution 1:2000; Santa Cruz Biotechnology Inc., USA) or mouse monoclonal anti-human vimentin (dilution 1:8000; Dako, Japan) overnight at 4ºC. Finally, the sections were incubated with secondary antibody (Histofine Simple Stain MAX-PO/MULTI; Nichirei, Japan) for 30 min at RT after rinsing with 0.01 M phosphate-buffered saline (PBS). The obtained products were observed with 3, 3'-Diaminobenzidine Tetrahydrochloride (DAB +, Liquid; Dako, Japan). Hematoxylin was used as the counterstain.

Four of 100 sections per subject were randomly selected, and IHC staining was performed with the same antibody on 3 of 4 sections. One section, used as the negative control, was incubated with normal mouse serum, diluted at the same concentration as the primary antibody. Renal biopsy sections from patients with membranous nephropathy were used as positive controls. All the sections were observed using a light microscope.

Quantitative analysis of PCNA, ICAM-1, and vimentin expression

The number of PCNA-, ICAM-1– and vimentin-positive LECs was counted in each section. Positive cell rate was defined as the percentage of cells that showed expression of the antigens in any position of the cell membrane. The average positive cell rate obtained from 3 sections per stain was used for statistical analysis.

Immunolocalization of ICAM-1

ICAM-1 was immunolocalized on the surface, side, and basement of LECs. The study subject was considered as positive if the average positive cell rate of ICAM-1 expression in the sections analyzed was > 25%. The ratio of the number of positive subjects to the total number of subjects from 3 immunolocalization findings of ICAM-1 was obtained.

Statistical analysis

The Statistical Package for Social Sciences for Windows (SPSS Inc., PASW Statistics, version 18) was used for statistical analysis, and findings were considered to be statistically significant if $P < 0.05$. The differences in the expression of PCNA, ICAM-1, vimentin, the severity of different types of cataracts, age, and the immunolocalization findings of ICAM-1 in cataract LECs between the DM and control groups were analyzed by Student's *t*-test, Mann-Whitney *U*-test, and Pearson's Chi-square test, as appropriate. The correlations between PCNA, ICAM-1 expression and clinical parameters in the DM group were analyzed by Spearman's rank correlation test. The correlations among ICAM-1, PCNA, and vimentin expression were analyzed by Spearman's rank correlation test. The receiver operator characteristic (ROC) curve was used to determine the optimal cut-off value of ICAM-1 expressed on the side and basement of LECs, according to the coordinates of the cut-off point of the ROC curve that was closest to the coordinate (0, 1). The odds ratio of the immunolocalization of ICAM-1 on the surface, side, and basement of LECs was also evaluated as a risk factor associated with the progression of cataract in type 2 diabetes between the DM and the control groups.

RESULTS

Clinical characteristics

Patient characteristics are summarized in Table 1. The classification of cataract, according to the LOCS Ⅲ criteria, in DM and control groups is shown in Table 2 [12]. No significant difference in the severity

Table 1 Patient characteristics and results of immunohistochemistry staining

	DM group	Control group
Number of patients	25	25
Age (years; mean ± SD)	67.88 ± 16.26	70.84 ± 7.89
Gender (Male/Female)	14/11	10/15
HbA1c (%; mean ± SD)	6.72 ± 1.04	5.01 ± 0.98
Duration of diabetes (years; mean ± SD)	15.86 ± 15.59	0
Positive cell rate of PCNA (%; mean ± SD)	66.97 ± 15.92	77.75 ± 15.24 *
Positive cell rate of ICAM-1 (%; mean ± SD)	74.89 ± 18.16	38.30 ± 16.85 **
Positive cell rate of vimentin (%; mean ± SD)	95.65 ± 3.58	91.41 ± 12.02
ICAM-1 on the surface of LECs (positive subject ratio)	24/25	20/25
ICAM-1 on the side of LECs (positive subject ratio)	24/25	16/25 ***
ICAM-1 on the basement of LECs (positive subject ratio)	18/25	3/25 ****

* ($P < 0.05$), DM group vs. control group.
** ($P < 0.0001$), DM group vs. control group.
*** ($P < 0.01$), DM group vs. control group.
**** ($P < 0.0001$), DM group vs. control group.
DM: diabetes mellitus; HbA1c: hemoglobin A1c; PCNA: proliferating cell nuclear antigen; ICAM-1: intercellular adhesion molecule-1; LECs: lens epithelial cells

Table 2 Classifications of cataracts in diabetes mellitus and control groups, according to the Lens Opacities Classification System Ⅲ criteria

Grade		0	1	2	3	4	5
Cortical opacity	DM	11/25	5/25	5/25	2/25	2/25	0/25
(positive subjects/total subjects)	Control	7/25	5/25	1/25	4/25	8/25	0/25
Nuclear opacity	DM	4/25	0/25	12/25	8/25	1/25	0/25
(positive subjects/total subjects)	Control	2/25	1/25	14/25	6/25	2/25	0/25
Posterior subcapsular opacity	DM	10/25	10/25	1/25	3/25	1/25	0/25
(positive subjects/total subjects)	Control	16/25	2/25	3/25	1/25	2/25	1/25

DM: diabetes mellitus

of different types of cataracts, i.e., cortical, nuclear, and posterior subcapsular cataract ($P = 0.065$, $P = 0.924$, and $P = 0.347$, respectively) and age ($P = 0.756$) between the DM and control groups was noted. Almost all the subjects under study had complex cataracts, with more than 2 kinds of opacities occurring concurrently.

PCNA expression in LECs

The positive cell rates of PCNA expression in the DM and control groups were 66.97% ± 15.92% and 77.75% ± 15.24%, respectively. PCNA antigen level was higher in the control group than in the DM group ($P = 0.036$; Fig. 1, Table 1).

ICAM-1 expression in LECs

The positive cell rate of ICAM-1 expression was 74.89% in the DM group and 38.30% in the control group. The level of ICAM-1 expression was significantly different between the 2 groups ($P < 0.0001$, Table 1). ICAM-1 was immunolocalized on the surface, side, and basement of LECs (Fig. 1). In the DM group, ICAM-1 expression was observed on each side of several LECs, while in the control group, it was mainly observed on the surface and partly on the side of LECs. The ratio of the number of positive subjects with ICAM-1 immunolocalization on the surface, side, and basement of LECs to the total number of subjects in the 2 groups is shown in Table 1. Compared to that in the control group, a significant increase in ICAM-1 expression (positive subject ratio) was found in the DM group, and this finding was consistent with ICAM-1 immunolocalization on the side and basement of LECs ($P = 0.005$ and $P < 0.0001$, respectively). However, no difference was found in ICAM-1 expression on the surface of LECs ($P = 0.082$).

Vimentin expression in LECs

Vimentin was expressed in almost all the cells in both DM and control groups (Fig. 1). No significant difference was noted in the positive cell rates between the 2 groups (95.65% ± 3.58% in the DM group and 91.41% ± 12.02% in the control group; $P = 0.264$; Table 1).

Correlations between PCNA, ICAM-1 expression in LECs and several clinical and immunohistochemical parameters

In the DM group, no significant correlation was noted between ICAM-1 expression and several clinical parameters, such as age, duration of diabetes, HbA1c level, severity of DR, and severity of different types of cataracts (Table 3); PCNA expression had a negative correlation with duration of diabetes ($r = -0.478$, $P < 0.05$) but a positive correlation with subcapsular opacity ($r = 0.661$, $P < 0.001$; Table 3). A negative correlation was found between PCNA and ICAM-1 expression ($r = -0.561$, $P < 0.0001$), but no correlation was found between PCNA and vimentin expression ($r = 0.151$, $P = 0.341$).

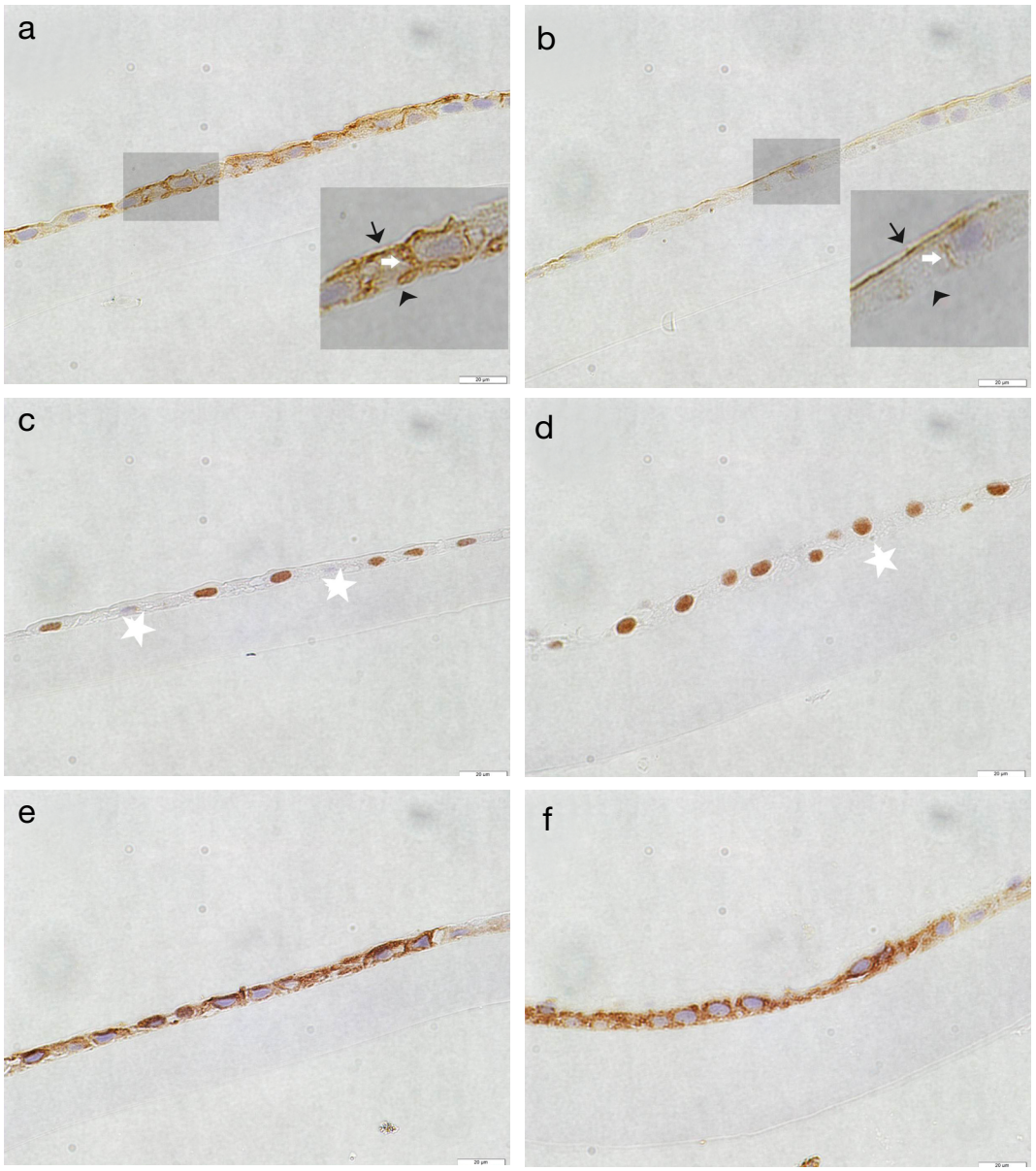

Fig. 1 **a, b** Expression of intercellular adhesion molecule-1 (ICAM-1) in the lens epithelial cells (LECs). ICAM-1 was expressed on the surface (black arrow), side (white arrow), and basement (black arrowhead) of LECs. In the DM group **(a)**, ICAM-1 expression was observed on every side of several LECs, while in the control group **(b)**, it was expressed mainly on the surface and partly on the side of LECs. **c, d** Expression of proliferating cell nuclear antigen (PCNA) in LECs. PCNA expression in LECs in the DM group **(c)** is lower than that in the control group **(d)**, in which negative PCNA expression is marked by an asterisk in white. **e, f** Expression of vimentin in LECs. The vimentin expression in LECs is not significantly different between the DM group **(e)** and the control group **(f)**. Immunohistochemistry staining; Scale Bar = 20 μ m.

ROC curve analysis and odds ratio of ICAM-1 expression in LECs

The ROC curve was analyzed, and the optimal cut-off values of ICAM-1 expression on the side and basement of LECs were 35.90% and 63.83%, respectively. Immunolocalization of ICAM-1 on the surface of LECs did not differ significantly (odds ratio = 6.000; 95% confidence interval, 0.647–55.661) between the 2 groups, while the odds ratios of the immunolocalization of ICAM-1 on the side and basement of LECs were higher in the DM group (13.500; 95% confidence interval, 1.556–117.137 and 18.857; 95% confidence interval, 4.254–83.592, respectively) than in the control group.

DISCUSSION

Studies on both animals and humans have highlighted the important contribution of increased AR activity, which can lead to multiple metabolic and signal-transduction changes, ultimately affecting transcriptional regulation and gene expression in LECs subject to diabetic cataract [1]. Several factors have been identified as risk factors for the onset of diabetic cataract [1]. Especially in elderly subjects, diabetic cataract onset has been shown to be associated with age, increased severity of retinopathy, diuretic usage, low intraocular pressure, smoking, and low diastolic blood pressure but not with duration of diabetes and

Table 3 Results of the correlation analysis to assess factors relevant to the expression of proliferating cell nuclear antigen (PCNA) and intercellular adhesion molecule-1 (ICAM-1) in lens epithelial cells in patients with type 2 diabetes

	PCNA		ICAM-1	
	r	*P* value	*r*	*P* value
Age	0.049	0.816	0.034	0.871
Duration of diabetes	- 0.478	0.016*	0.261	0.208
HbA1c level	- 0.153	0.465	- 0.067	0.752
Diabetic retinopathy	0.152	0.468	- 0.035	0.868
Cortical cataract	- 0.055	0.795	0.152	0.469
Nuclear cataract	- 0.228	0.272	- 0.054	0.797
Posterior subcapsular cataract	0.661	0.000**	- 0.250	0.229

P value: correlation is significant if $P < 0.05$ (2-tailed).
*: $P < 0.05$; **: $P < 0.001$.
r: regression coefficient.

glycosylated hemoglobin levels. In our study, there was no difference in the age and severity of the different types of cataracts between the 2 groups (Table 1).

LECs can undergo proliferation, migration, transdifferentiation, or apoptosis from cataract onset to maturation in different types of cataracts [14–17]. Bras *et al.* showed that there was a higher expression of PCNA in LECs from cataract patients with diabetes compared with normal patients and those with inherited cataracts, but they found no significant difference in DNA damage (Gadd45) or cell apoptosis in LECs from diabetic dogs with anterior subcapsular cataracts [18]. In our study performed on cataract patients without anterior subcapsular opacities, the PCNA expression in DM group had a positive correlation with subcapsular opacity, which suggested that the increased PCNA expression maybe aggravate the subcapsular opacity. PCNA was expressed in 77.75% of LECs in the control group and 65.08% of LECs in the DM group; its expression in the DM group had a negative correlation with duration of diabetes, indicating the decreased proliferation ability of LECs as a risk factor for the progression of human cataract with type 2 diabetes (Fig. 1, Table 1).

In all kinds of ocular pathological situations, such as disciform herpes simplex virus (HSV) keratitis [19], allergic conjunctivitis [20], uveitis [21] and epiretinal membrane [22], ICAM-1, also known as an inflammatory molecule, is considered necessary for cell adhesion, migration, proliferation, apoptosis, and cell signal transmission. Inflammatory cytokines, such as interleukin (IL)-1 [23] and tumor necrosis factor (TNF)-alpha [24] have increased the expression of ICAM-1 on multiple cell types. Nishi *et al.* indicated that cell adhesion molecules, including ICAM-1, may serve in the attachment process of cataractous LECs to extracellular matrix and may be involved in the formation and disruption of cell-to-cell and cell-to-posterior capsule interactions when LECs migrate onto the posterior capsule after surgery [7]. Consequently, we have paid special attention to the expression of ICAM-1 in LECs. In this study, we first reported the immunolocalization of ICAM-1 on the surface, side, and basement of LECs (Fig. 1). ICAM-1 was expressed only on the surface of LECs when the positive cell rate was < 35.90%. With an increase in the level of ICAM-1 expression (positive cell rate range, 35.90–63.83%), the protein was detected both on the surface and side of LECs. Finally, when the expression of ICAM-1 was higher than a certain level (positive cell rate, > 63.83%), the expression of ICAM-1 also extended from the side to the basement of LECs, especially in the DM group. Moreover, the odds ratios of the immunolocalization of ICAM-1 on the side and basement of LECs were 13.500-fold and 18.857-fold higher in the DM group, respectively, than in the control group. Thus, ICAM-1 may be an important risk factor during the progression of cataract in type 2 diabetic patients.

Tanaka *et al.* have demonstrated that ICAM-1–positive synovial cells were growth-arrested and underwent subsequent apoptosis, whereas ICAM-1–negative synovial cells were proliferative [25]. In the present study, PCNA expression was negatively correlated with ICAM-1 expression in all subjects, which indicated that during the progression of cataract, the proliferation ability of LECs is decreased when there is an over expression of ICAM-1. Therefore, it is possible that ICAM-1 expression and cell proliferation in the lens epithelium of cataract are correlated. Further research is needed to identify such relationships between ICAM-1 expression and proliferation in cataractous LECs in order to clarify cataract pathogenesis, especially in diabetes patients.

ACKNOWLEDGMENTS

We thank the members in the Department of Histological Science of Tokai University School of Medicine for their excellent technical assistance. We also thank all the members in the Department of Ophthalmology of Tokai University School of Medicine who provided the materials for the study.

REFERENCES

1) Obrosova IG, Chung SS, Kador PF. Diabetic cataracts: mechanisms and management. Diabetes Metab Res Rev 2010; 26:172–80.
2) Gonzalez K, McVey S, Cunnick J, Udovichenko IP, Takemoto DJ. Acridine orange differential staining of total DNA and RNA in normal and galactosemic lens epithelial cells in culture using flow cytometry. Curr Eye Res 1995; 14: 269–73.
3) Murata M, Ohta N, Sakurai S, Alam S, Tsai J, Kador PF, *et al.* The role of aldose reductase in sugar cataract formation: aldose reductase plays a key role in lens epithelial cell death (apoptosis). Chem Biol Interact 2001; 130–132(1–3): 617–25.
4) Takamura Y, Kubo E, Tsuzuki S, Akagi Y. Apoptotic cell death in the lens epithelium of rat sugar cataract. Exp Eye Res 2003; 77:

51–57.
5) Vorkauf M, Duncker G, Nölle B, Sterry W. Adhesion molecules in normal human conjunctiva. An immunohistological study using monoclonal antibodies. Graefes Arch Clin Exp Ophthalmol 1993; 231: 323–30.
6) Barile GR, Chang SS, Park LS, Reppucci VS, Schiff WM, Schmidt AM. Soluble cellular adhesion molecules in proliferative vitreoretinopathy and proliferative diabetic retinopathy. Curr Eye Res 1999; 19: 219–27.
7) Nishi O, Nishi K, Akaishi T, Shirasawa E. Detection of cell adhesion molecules in lens epithelial cells of human cataracts. Invest Ophthalmol Vis Sci 1997; 38: 579–85.
8) Glushakova O, Kosugi T, Roncal C, Mu W, Heinig M, Cirillo P, *et al.* Fructose induces the inflammatory molecule ICAM-1 in endothelial cells. J Am Soc Nephrol 2008; 19: 1712–20.
9) Mattioli LF, Holloway NB, Thomas JH, Wood JG. Fructose, but not dextrose, induces leukocyte adherence to the mesenteric venule of the rat by oxidative stress. Pediatr Res 2010; 67: 352–56.
10) Raghu Kalluri and Eric G. Neilson. Epithelial-mesenchymal transition and its implications for fibrosis. J Clin Invest 2003; 112(12): 1776–84.
11) Joo CK, Lee EH, Kim JC, Kim YH, Lee JH, Kim JT, *et al.* Degeneration and transdifferentiation of human lens epithelial cells in nuclear and anterior polar cataracts. J Cataract Refract Surg 1999; 25: 652–58.
12) Chylack LT Jr, Wolfe JK, Singer DM, Leske MC, Bullimore MA, Bailey IL, *et al.* The Lens Opacities Classification System Ⅲ. The Longitudinal Study of Cataract Study Group. Arch Ophthalmol 1993; 111: 831–36.
13) Report of the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Diabetes Care 1997; 20: 1183–97.
14) Zemaitiene R. Posterior capsule opacification: incidence and pathogenesis. Medicina (Kaunas) 2003; 39: 830–37.
15) Ye L, Li Q, Cai X, Deng P. Expression of TGF-beta2 mRNA and PCNA, FN protein in lens epithelial cells in age-related nuclear and cortex cataract. J Huazhong Univ Sci Technolog Med Sci 2005; 25: 578–81.
16) Li WC, Kuszak JR, Dunn K, Wang RR, Ma W, Wang GM, *et al.* Lens epithelial cell apoptosis appears to be a common cellular basis for non-congenital cataract development in humans and animals. J Cell Biol 1995; 130: 169–81.
17) Harocopos GJ, Alvares KM, Kolker AE, Beebe DC. Human age-related cataract and lens epithelial cell death. Invest Ophthalmol Vis Sci 1998; 39: 2696–06.
18) Bras ID, Colitz CM, Kusewitt DF, Chandler H, Lu P, Gemensky-Metzler AJ, *et al.* Evaluation of advanced glycation end-products in diabetic and inherited canine cataracts. Graefes Arch Clin Exp Ophthalmol 2007; 245: 249–57.
19) Elner VM, Dutt S, Pavilack MA, Sugar A, Foster CS, Elner SG. Intercellular adhesion molecule-1 (ICAM-1) and HLA-DR antigens in herpes keratitis. Ophthalmology 1992; 99: 1400–07.
20) Ciprandi G, Buscaglia S, Pesce G, Villaggio B, Bagnasco M, Canonica GW. Allergic subjects express intercellular adhesion molecule-1 (ICAM-1 or CD54) on epithelial cells of conjunctiva after allergen challenge. J Allergy Clin Immunol 1993; 91: 783–92.
21) Wakefield D, McCluskey P, Palladinetti P. Distribution of lymphocytes and cell adhesion molecules in iris biopsy specimens from patients with uveitis. Arch Ophthalmol 1992; 110: 121–25.
22) Heidenkummer HP, Kampik A. Kampik. Intercellular adhesion molecule-1 (ICAM-1) and leukocyte function-associated antigen-1 (LFA-1) expression in human epiretinal membranes. Graefes Arch Clin Exp Ophthalmol 1992; 230: 483–87.
23) Dustin ML, Rothlein R, Bhan AK, Dinarello CA, Springer TA. Induction by IL 1 and interferon-gamma: tissue distribution, biochemistry, and function of a natural adherence molecule (ICAM-1). J Immunol 1986; 137: 245–54.
24) Pober JS, Bevilacqua MP, Mendrick DL, Lapierre LA, Fiers W, Gimbrone MA Jr. Two distinct monokines, interleukin 1 and tumor necrosis factor, each independently induce biosynthesis and transient expression of the same antigen on the surface of cultured human vascular endothelial cells. J Immunol 1986; 136: 1680–87.
25) Tanaka Y, Nomi M, Fujii K, Hübscher S, Maruo A, Matsumoto S, *et al.* Intercellular adhesion molecule 1 underlies the functional heterogeneity of synovial cells in patients with rheumatoid arthritis: involvement of cell cycle machinery. Arthritis Rheum 2000; 43: 2513–22.