Jpn J Ophthalmol 2011;55:22–27
DOI 10.1007/s10384-010-0904-z

CLINICAL INVESTIGATION

Changes to Upper Eyelid Orbital Fat from Use of Topical Bimatoprost, Travoprost, and Latanoprost

Juwan Park, Hyun Kyung Cho, and Jung-Il Moon

Department of Ophthalmology and Visual Science, St. Mary's Hospital, The Catholic University of Korea, Seoul, Korea

Abstract

Purpose: To confirm the possible mechanism by which topical prostaglandin antiglaucoma drugs cause a deep superior sulcus.

Methods: Among patients who used bimatoprost (Lumigan), latanoprost (Xalatan), or travoprost (Travatan) and who developed a deep upper lid sulcus, 18 eyes of 11 patients (mean age, 58.2 ± 8.9 years) were studied. Seven patients were binocular users of one of the eye drops and four were monocular users. Preaponeurotic orbital fat was obtained, and the mean adipocyte density compared.

Results: In the four monocular users, mean adipocyte density of treated eyes was 1758.21 ± 158.15 cells/mm^2, and that of untreated eyes was 1258.73 ± 127.54 cells/mm^2. This difference was statistically significant ($P = 0.04$), suggesting that the adipocytes were atrophied in the treated eyes. The mean adipocyte density of the bimatoprost group was 2073.35 ± 184.89 cells/mm^2, that of the travoprost group was 1623.46 ± 218.99 cells/mm^2, and that of the latanoprost group was 1468.20 ± 113.44 cells/mm^2. The densities of the bimatoprost and travoprost groups, but not of the latanoprost group ($P = 0.75$), were significantly different from that of the untreated group ($P < 0.001$).

Conclusions: Fat atrophy can be considered a mechanism of upper eyelid sulcus deepening in patients using topical prostaglandin analogs. **Jpn J Ophthalmol** 2011;55:22–27 © Japanese Ophthalmological Society 2011

Keywords: bimatoprost, deep superior sulcus, latanoprost, orbital fat atrophy, travoprost

Introduction

Prostaglandin F2α (PGF2α) analogs such as latanoprost (Xalatan, Pharmacia, Peapack, NJ, USA) and travoprost (Travatan, Alcon, Fort Worth, TX, USA), and also similar prostamide (prostaglandin ethanolamide) analogs, that is, 17-phenyl-derivatives of prostamide F2α such as bimatoprost (Lumigan, Allergan, Irvine, CA, USA), are topical hypotensive drugs frequently used for treating ocular hypertension and glaucoma.[1–4]

These drugs are well known for their efficacy and potency, as well as for good patient compliance because they can be administered in a single daily dose. However, the use of these drugs may lead to adverse effects such as darkening of the periocular skin and iris, increased eyelash growth, cystoid macular edema, and conjunctival hyperemia.[5–7]

Deepening of the upper lid sulcus and decreased dermatochalasis have been recently reported in long-term bimatoprost (Lumigan) users, and some of these side effects are reported to be reversible once the drug is stopped.[8–12] Travoprost (Travatan) is reported to induce deepening of the upper lid sulcus with long-term use, a condition reversed by discontinuation of the medication.[13] We have also observed these side effects, finding these changes not only in bimatoprost and travoprost users but also in some long-term users of latanoprost.

Different mechanisms have been suggested to explain the different adverse effects of topical prostaglandin analogs. A few recent reports postulated that periocular skin/iris darkening, cystoid macular edema, and conjunctival

Received: February 22, 2010 / Accepted: September 28, 2010
Correspondence and reprint requests to: Jung-Il Moon, Department of Ophthalmology and Visual Science, St. Mary's Hospital, The Catholic University of Korea, #62 Yeouido-dong, Yeongdeungpo-gu, Seoul 150-713, Korea
e-mail: jimoon@catholic.ac.kr

hyperemia are related to the actions of prostaglandins associated with stimulation of melanogenesis in the skin or iris, disruption of the blood–aqueous barrier in pseudophakes, and the release of nitric oxide.[5–7]

However, all of these proposed mechanisms for the known adverse effects of bimatoprost and other prostaglandin analogs do not appear to cause deepening of the lid sulcus. It has been postulated that either Műller muscle contraction or fibrosis and orbital fat atrophy may affect the development of this adverse effect,[9] but the exact mechanism remains unknown and further investigation of this new adverse effect is necessary.

In the present study, we focused on changes in orbital fat to investigate the possible mechanism of this adverse effect, which is reported to be reversible to some degree. We biopsied the preaponeurotic orbital fat in monocular and binocular bimatoprost, travoprost, and latanoprost users who had developed a deep superior sulcus. The adipocyte density and the other pathologic changes were then histologically inspected to confirm the orbital fat changes in users of any of these three drugs.

Material and Methods

Study Population

Among patients using bimatoprost (Lumigan), latanoprost (Xalatan), or travopost (Travatan) for ocular hypertension and glaucoma and who developed a deep upper lid sulcus, 11 (six men and five women; mean age, 58.2 ± 8.9 years; 18 eyes) agreed to participate in the present study and their signed informed consent was obtained. We analyzed the adipocyte density of the preaponeurotic orbital fat of the upper lids of the 18 eyes. Seven patients were using one of the three types of eye drops in both eyes, and four were using eye drops in only one eye. Among the four monocular users, two were using latanoprost and the other two were using bimatoprost or travoprost. The mean duration of eye-drop usage was 4.8 ± 1.7 years; some of the patients were also using hypotensive eye drops of another type (Table 1).

Patients were excluded if they had a preexisting deep upper lid sulcus, determined from a photograph taken before eye-drop use, or preexisting blepharoptosis causing a deep superior sulcus. Patients who had a history of periocular trauma or surgery were also excluded.

The present study was approved by the institutional review board of St. Mary's Hospital, College of Medicine, the Catholic University of Korea.

Preparation of the Orbital Fat

The biopsy procedure for preaponeurotic orbital fat of the upper lid was performed under local anesthesia by one ophthalmic plastic surgeon (J.P.). Lidocaine mixed with 1:100 000 epinephrine was injected subcutaneously into the upper lid, and a skin incision 5 mm in length was made in the center of the upper lid crease. Preaponeurotic orbital fat was obtained through the incision made for the biopsy. To avoid "crush" injury and electric damage to the fat tissue, a #15 knife was used to resect the fat tissue instead of scissors and electrocautery. During the procedure, compression of the fat tissue was also avoided, and minimal, gentle handling was done. Hemostasis with electrocautery was not performed until the incision biopsy of the fat tissue was completed. The skin was sutured with #6-0 Prolene, and the stitches were taken out 5 days after the procedure.

Measurement of Mean Adipocyte Density and the Pathologic Study

To see if there were any errors or differences between the two fixation methods, both frozen sections and permanent sections were made of tissue from the first three patients enrolled in the study (six eyes), and mean adipocyte density was compared between the two types of sections. The results showed no statistically significant differences between the two fixation methods. After this, frozen sections were not made, and the biopsied fat tissue was immediately soaked in 10% formalin solution. The fixed fat tissue was stained

Table 1. Characteristics of the patients

	Treated group			
	Bimatoprost (Lumigan)	Travoprost (Travatan)	Latanoprost (Xalatan)	Untreated group
Number of eyes	5	5	8	4
Number of patients (binocular users/monocular users)	3 (2/1)	3 (2/1)	5 (3/2)	
Mean age (years), mean ± SD	58.6 ± 10.3	61.3 ± 9.8	57.9 ± 7.5	59.3 ± 6.4
Sex (M:F)	1:2	2:1	3:2	2:2
Duration of eye-drop usage (years), mean ± SD*	2.4 ± 0.8	4.8 ± 2.3	5.9 ± 3.6	—
Cosopt co-use (eyes)	—	—	2	—
Cosopt & Alphgan P co-use (eyes)	3	4	3	—

Age and sex did not differ significantly among the groups (age, $P = 0.76$; sex, $P = 0.35$).
*$P = 0.02$.

with hematoxylin and eosin and observed under a light microscope.

For the measurement of adipocyte density, at least five nonoverlapping fields per specimen were analyzed at a magnification of ×100. The estimated adipocyte density was calculated as the number of adipocytes per unit area. When measuring the number of adipocytes, we avoided the periphery of the specimen, and blood vessels and connective tissue were not included in the unit area. The results of all histological analyses were averaged per specimen and per group, and the mean number of adipocytes per unit area was compared.

In addition to the measurement of the adipocyte density, to determine if there were other pathologic changes, we also had an experienced pathologist examine the specimens.

Data Analysis

SPSS 13.0 software was employed to analyze the data. All results are presented as means ± SD. Because the number of specimens was not high enough to use parametric tests, differences in mean adipocyte density between two groups (e.g., the treated eye and the untreated eye of the monocular users) were analyzed using the Mann-Whitney *U* test, and differences in mean adipocyte density among more than two groups (e.g., the bimatoprost-treated, latanoprost-treated, travoprost-treated, and untreated groups) were assessed using the Kruskall-Wallis test. Differences were considered statistically significant when *P* values were less than 0.05.

Results

Comparison of Adipocyte Density Between Frozen Sections and Permanent Sections

To determine the differences in cell density between the fixation methods, we made both frozen sections and permanent sections of the preaponeurotic orbital fat tissue obtained from the upper lids of the six eyes of the first three enrolled patients, and compared the adipocyte density between them. The mean adipocyte density of the frozen sections was 1831.45 ± 112.12 cells/mm^2, and that of the permanent sections was 1793.23 ± 112.12 cells/mm^2, a nonsignificant difference ($P = 0.76$, Wilcoxon signed-rank test). Therefore, for the additional patients, we made only permanent sections for measuring the mean adipocyte density.

Binocular Comparison of Adipocyte Density in Monocular Prostaglandin Users

The mean adipocyte density of the preaponeurotic orbital fat of the upper lids was compared between the two eyes of each of the four monocular users: two latanoprost users, one travoprost user, and one bimatoprost user. The mean adipocyte density of the treated eyes was 1758.21 ± 158.15 cells/mm^2, and that of the untreated eyes was 1258.73 ± 127.54 cells/mm^2, a significant difference ($P = 0.04$, Mann-Whitney *U*-test), suggesting that the adipocytes were atrophied in the treated eyes (Fig. 1).

Comparison of Adipocyte Density among the Bimatoprost-Treated, Latanoprost-Treated, Travoprost-Treated, and the Untreated Groups

To investigate the differences among eyes treated with one of the three different prostaglandins and untreated eyes, we used the measured adipocyte density values in eyes of both the binocular and monocular users.

The mean adipocyte density of the bimatoprost group was 2073.35 ± 184.89 cells/mm^2, that of the travoprost group was 1623.46 ± 218.99 cells/mm^2, and that of the latanoprost group was 1468.20 ± 113.44 cells/mm^2. These densities were all higher than those of the untreated eyes of the monocular users, and the differences among the four groups were statistically significant ($P = 0.03$, Kruskall-Wallis test).

Cell density in the bimatoprost group was significantly different from that in the travoprost, latanoprost, or untreated groups ($P < 0.001$, Mann-Whitney U-test). The travoprost and latanoprost groups were not significantly different from each other ($P = 0.672$, Mann-Whitney *U* test), yet a statistically significant difference existed between the travoprost group and the untreated group ($P < 0.001$, Mann-Whitney *U* test). The latanoprost group was not significantly different from the untreated group ($P = 0.75$, Mann-Whitney *U* test).

Other Pathologic Findings

Pathologic findings suggestive of fat atrophy, such as clumped nuclei of the adipocytes, were observed in the treated eyes (Fig. 2), but other pathologic changes such as inflammation, fibrosis, or necrosis of adipocytes were not detected.

Discussion

New periocular changes associated with topical prostaglandin therapy with widely used hypotensive drugs have recently been reported. Casson and Selva[14] described a patient whose trichomegaly secondary to the chronic use of latanoprost resulted in eyelash ptosis that obstructed his visual field and required a bilateral eyelid anterior lamellar transposition procedure.

Since Peplinski and Albiani Smith[8] first reported upper eyelid sulcus deepening and dermatochalasis involution in three Caucasian patients treated with bimatoprost unilaterally, other similar clinical observations have been reported. Yam et al.[12] described bilateral upper eyelid sulcus deepening and involution of dermatochalasis in an Asian patient

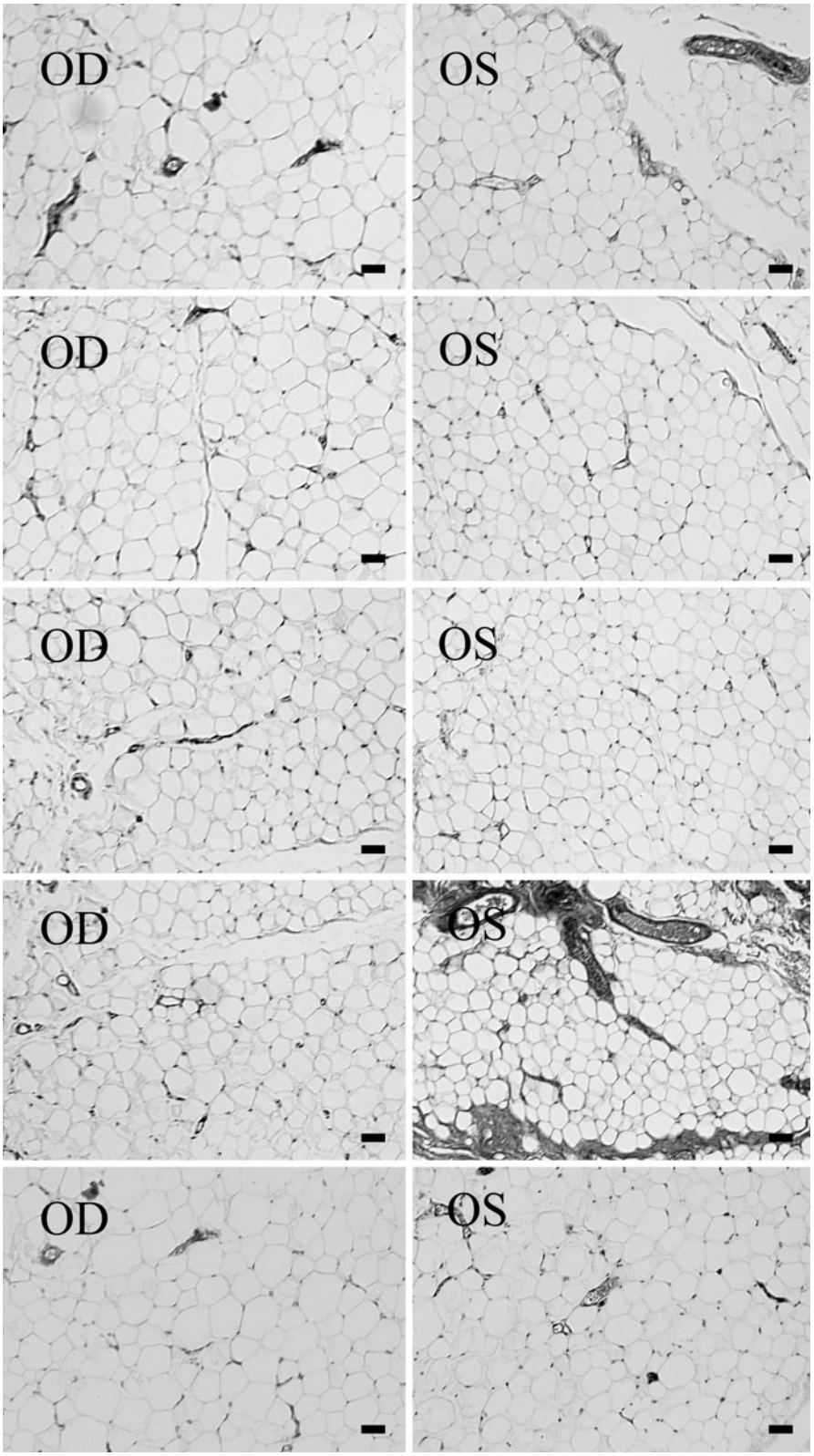

Figure 1. Microscopic findings of five different biopsy sites of the preaponeurotic orbital fat of a monocular bimatoprost user. The cell density of the fat tissue is higher in the treated eye (OS) than in the untreated eye (OD) (×100, H&E stain). Bars = 50 μm.

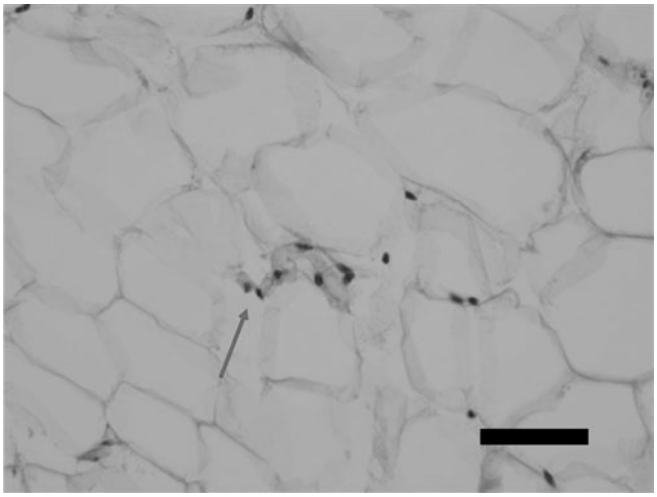

Figure 2. Microscopic findings showing atrophied fat tissue in a bimatoprost-treated eye. The *arrow* indicates clumped adipocyte nuclei, which implies fat atrophy (×200, H&E stain). Bar = 50 μm.

using bimatoprost bilaterally. Filippopoulos et al.[10] and Tappeiner et al.[11] reported the additional finding of enophthalmos. Recently, Yang et al.[13] reported unilateral deepening of the upper eyelid sulcus in two Asian patients treated with travoprost unilaterally that was reversed by discontinuation of the medication.

Other general conditions that cause a deep superior sulcus should be identified before a deep superior sulcus can be regarded as a side effect of topical prostaglandin use. Besides senile enophthalmos associated with aging,[15] an acquired deep superior sulcus is typically found in patients with involutional blepharoptosis as a result of levator aponeurosis dehiscence or disinsertion. But aponeurotic blepharoptosis is usually accompanied by not only a deep superior sulcus but also lid drooping and compensatory brow elevation, all of which are basically irreversible, so surgery is required for correction.

As a possible mechanism of upper eyelid sulcus deepening in prostaglandin analog users, Peplinski and Smith[8] suggested that bimatoprost may affect the Műller muscle. However, if either atrophy or fibrosis of the Műller muscle leads to upper eyelid sulcus deepening, then widening of the palpebral fissure should also happen. This change was not observed in our study; moreover, in most patients the upper eyelid sulcus deepening was reversible, which diminishes the possibility of Műller muscle fibrosis being the cause of the upper eyelid sulcus deepening.

Filippopoulos et al.[10] and Tappeiner et al.,[11] who demonstrated not only upper eyelid sulcus deepening but also reduced infraocular fat and enophthalmos, hypothesized that the common pathophysiologic mechanism of these periocular changes is atrophy of preaponeurotic and deep orbital fat.

In many patients, upper eyelid sulcus deepening is partially or completely reversible, and in some patients enophthalmos accompanies the upper eyelid sulcus deepening. Therefore, we inferred that changes in the orbital fat might be related to upper eyelid sulcus deepening and investigated histological changes of the orbital fat.

In fat atrophy, as opposed to pathologic fat tissue change in adults, the change of fat volume is controlled by the intracellular lipid content of the adipocytes rather than by the number of adipocytes. If the changes of the upper lid are caused by fat atrophy, then the mean number of adipocytes per unit area, that is, mean adipocyte density, should be increased by the reduced intracellular lipid content of the adipocytes. If subclinical chronic inflammation is the cause of the periocular changes, then it should be easy to identify it by pathologic examination. Therefore, to observe the changes in the orbital fat tissue, we evaluated adipocyte density as well as other pathologic changes such as inflammation or fibrosis, apoptosis, or necrosis by hematoxylin and eosin staining and light microscopy.

To the best of our knowledge, no previous studies have confirmed histological changes in prostaglandin analog users by orbital fat tissue biopsy. Moreover, in addition to bimatoprost users, the present study included users of two other prostaglandin analogs, and we observed a deep superior sulcus in all three prostaglandin analog groups. We compared the orbital fat tissue between these three groups and the untreated eyes of the monocular prostaglandin analog users. Our results revealed that the mean adipocyte density in the three prostaglandin analog-treated eyes was higher than that in the untreated eyes. We also observed such histological findings as clumped nuclei, which are suggestive of fat atrophy. Although the number of samples was small and the statistical significance was not strong, the adipocyte density was significantly different between the prostaglandin analog-treated eyes and the untreated eyes or among the three prostaglandin analogs; the mean adipocyte density was highest in the bimatoprost group followed in order by the travoprost and latanoprost groups.

The inhibiting effect of PGF2a on adipocyte differentiation has been shown by previous studies.[16–18] Reginato et al.[18] showed that PGF2α, by combining with the cell surface FP receptor to activate mitogen-activated protein kinase, has an antiadipogenic effect, and that it inhibits a nuclear hormone receptor, peroxisome proliferator-activated receptor γ. As a result it blocks adipogenesis, the process by which orbital fibroblasts differentiate into adipocytes. This antiadipogenic effect of PGF2α is also observed in mature adipocytes, and it could probably also be applied as therapy for obesity and diabetes.[18] In contrast, prostaglandin J2 activates adipogenesis.

Although neither the exact receptors of bimatoprost nor its effect on fat tissue are yet clear, the present study results showed that, like PGF2α, bimatoprost also seems to have an antiadipogenic effect, presumed to be stronger than that of PGF2α.

Even though the small amount of biopsied specimen from the central preaponeurotic fat cannot represent the whole state of the orbital fat, and a two-dimensional adipocyte cell count cannot be used to assess the status of whole

adipocytes, the biopsies were performed in the same area of orbital fat tissue under the same conditions. The results revealed that the mean adipocyte density in the prostaglandin-treated eyes was significantly higher than that in untreated eyes. Therefore, fat atrophy is a possible mechanism for the development of a deep superior sulcus in topical prostaglandin users.

However, the number of samples was small: hence, the statistical significance was not strong. A prospective study with more samples needs to be done in the future. Because not all of the patients showed full recovery from their upper eyelid sulcus deepening after they stopped using the prostaglandin analogs, other, irreversible factors may be partly associated with this adverse effect. Therefore, further evaluation of the surrounding tissue, such as the Műller muscle, is required. An additional study employing cell culture techniques is necessary to overcome the limits of morphological inspection.

In conclusion, in the present study, we confirmed orbital fat atrophy to be a mechanism of upper eyelid sulcus deepening in topical prostaglandin analog users, including those using bimatoprost. When prescribing these drugs, this adverse effect should be explained to patients and monitoring for possible adverse effects should be performed.

References

1. Krauss, AH, Woodward DF. Update on the mechanism of action of bimatoprost: a review and discussion of new evidence. Surv Ophthalmol 2004;49:S5–11.
2. Hylton C, Robin AL. Update on prostaglandin analogs. Curr Opin Ophthalmol 2003;14:65–69.
3. Eisenberg DL, Toris CB, Camras CB. Bimatoprost and travoprost: a review of recent studies of two new glaucoma drugs. Surv Ophthalmol 2002;47:S105–115.
4. DuBiner H, Cooke D, Dirks M, Stewart WC, VanDenburgh AM, Felix C. Efficacy and safety of bimatoprost in patients with elevated intraocular pressure: a 30-day comparison with latanoprost. Surv Ophthalmol 2001;45:S353–360.
5. Miyake K, Ibaraki N. Prostaglandins and cystoid macular edema. Surv Ophthalmol 2002;47:S203–218.
6. Vogel R, Strahlman E, Rittenhouse KD. Adverse events associated with commonly used glaucoma drugs. Int Ophthalmol Clin 1999;39:107–124.
7. Easthope SE, Perry CM. Topical bimatoprost: a review of its use in open-angle glaucoma and ocular hypertension. Drug Aging 2002;19:231–248.
8. Peplinski LS, Albiani Smith K. Deepening of lid sulcus from topical bimatoprost therapy. Optom Vis Sci 2004;81:574–577.
9. Lee JW, Kim DY, Lee YK. Two cases of deepening of the upper lid sulcus from topical bimatoprost therapy. J Korean Ophthalmol Soc 2007;48:332–336.
10. Filippopoulos T, Paula JS, Torun N, Hatton MP, Pasquale LR, Grosskreutz CL. Periorbital changes associated with topical bimatoprost. Ophthal Plast Reconstr Surg 2008;24:302–307.
11. Tappeiner C, Perren B, Iliev ME, Frueh BE, Goldblum D. Orbital fat atrophy in glaucoma patients treated with topical bimatoprost—can bimatoprost cause enophthalmos? Klin Monbl Augenheilkd 2008;225:443–445.
12. Yam JC, Yuen NS, Chan CW. Bilateral deepening of upper lid sulcus from topical bimatoprost therapy. J Ocul Pharmacol Ther 2009;25:471–472.
13. Yang HK, Park KH, Kim TW, Kim DM. Deepening of eyelid superior sulcus during topical travoprost treatment. Jpn J Ophthalmol 2009;53:176–179.
14. Casson RJ, Selva D. Lash ptosis caused by latanoprost. Am J Ophthalmol 2005;139:932–933.
15. Rohrich RJ, Coberly DM, Fagien S, Stuzin JM. Current concepts in aesthetic upper blepharoplasty. Plast Reconstr Surg 2004;113: 32e–42e.
16. Serrero G, Lepak N. Endocrine and paracrine negative regulators of adipose differentiation. Int J Obes Relat Metab Disord 1996; 20:S58–64.
17. Serrero G, Lepak NM. Prostaglandin F2alpha receptor (FP receptor) agonists are potent adipose differentiation inhibitors for primary culture of adipocyte precursors in defined medium. Biochem Biophys Res Commun 1997;233:200–202.
18. Reginato MJ, Krakow SL, Bailey ST, Lazar MA. Prostaglandins promote and block adipogenesis through opposing effects on peroxisome proliferator-activated receptor gamma. J Biol Chem 1998;273:1855–1858.