REVIEW

Corneal Regeneration After Photorefractive Keratectomy: A Review

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Abstract Photorefractive keratectomy (PRK) remodels corneal stroma to compensate refractive errors. The removal of epithelium and the ablation of stroma provoke the disruption of corneal nerves and a release of several peptides from tears, epithelium, stroma and nerves. A myriad of cytokines, growth factors, and matrix metalloproteases participate in the process of corneal wound healing. Their balance will determine if reepithelization and stromal remodeling are appropriate. The final aim is to achieve corneal transparency for restoring corneal function, and a proper visual quality. Therefore, wound-healing response is critical for a successful refractive surgery. Our goal is to provide an overview into how corneal wounding develops following PRK. We will also review the influence of intraoperative application of mitomycin C, bandage contact lenses, anti-inflammatory and other drugs in preventing corneal haze and post-PRK pain.

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Keywords Photorefractive keratectomy; Cornea; Wound healing; Contact lenses

PALABRAS CLAVE
Queratectomía fotorrefractiva; Córnea; Curación de heridas; Lentes de contacto

Regeneración de la córnea tras queratectomía fotorreactiva: revisión bibliográfica

Resumen La queratectomía fotorreactiva (PRK) remodela el estroma de la córnea para compensar los errores refractivos. La eliminación del epitelio y la ablación del estroma provoca la alteración de los nervios corneales y la liberación de diversos péptidos de la lágrima, epitelio, estroma y nervios. Innumerables citoquinas, factores de crecimiento y metalloproteasas de la matriz participan en el proceso de regeneración y cicatrización corneal. Su equilibrio determinará si la re-epitelización y la remodelación del estroma son adecuados. El objetivo final...
el logro de la transparencia corneal para restablecer la función de la córnea, así como la calidad visual adecuada. Por tanto, la respuesta de regeneración y cicatrización corneal es esencial para el éxito de la cirugía refractiva. Nuestro objetivo es aportar una visión general sobre el modo en que se desarrolla dicho proceso tras la PRK. Revisaremos también la influencia de la aplicación intraoperatoria de mitomicina C, lentes de contacto terapéuticas, y otros fármacos para prevenir el haze y el dolor tras la PRK.

Las células epiteliales pueden mostrar apoptosis, una condición que resulta en la muerte celular programada. La apoptosis es un proceso crítico en el desarrollo, la diferenciación y la reparación del tejido corneal. En la cirugía refractiva, como la PRK, la apoptosis juega un papel importante en la cicatrización corneal.

**Epithelial Wound Healing Following PRK**

El epitelio corneal está formado por células superficiales, alas y lágrimas. En orden de facilitar la ablación del stroma en PRK, el epitelio corneal es removido. La ausencia del epitelio cambia la regeneración corneal. Epiteliales corneales son las primeras células involucradas en la regeneración del epitelio corneal después de PRK. Los epiteliales corneales proliferan y migran desde el limbo y el epitelio corneal basal para reestablecer la capa corneal. La regeneración del epitelio corneal después de PRK puede ser mejor entendida usando la actual confocal microscopy que se utiliza para visualizar la real estructura corneal. Se ha usado en animales y en humanos para visualizar la estructura corneal en tiempo real. Las observaciones sugieren que el número de células superficiales se redujo en el medio deshidratante comparado con condiciones normales, y el número de células corneales basales aumentó. Los estudios histológicos confirmaron que el epitelio corneal es más grueso después de PRK, causado por una elongación del epitelio corneal y un número aumentado de células corneales superficiales. El estiramiento y el engrosamiento de la córnea myópica PRK puede resultar en cicatrización corneal debido a los cambios en los tejidos como resultado de la ablación del stroma.

**Corneal Wound Healing**

El epitelio corneal es un epitelio queratinizado multilaminar en el que el epitelio superóptico proporciona la barrera externa. El epitelio corneal está compuesto por células estratificadas de diferentes tipos, con un alto contenido de queratina y una densidad celular de 50 000 células por cm². Las células corneales tienen un alto contenido de queratina y una alta densidad celular, lo que les permite resistir las condiciones ambientales adversas. Las células corneales también tienen una alta capacidad de proliferación y diferenciación, lo que les permite repararse después de lesiones.

La principal función de las células corneales es la protección de la córnea y la visión. Las células corneales también desempeñan un papel importante en la regulación del metabolismo y la homeostasis del tejido corneal. Las células corneales epiteliales tienen una alta capacidad de proliferación y diferenciación, lo que les permite repararse después de lesiones. Las células corneales epiteliales también liberan factores de crecimiento y factores de transcripción que promueven la regeneración del tejido corneal.

La regeneración del epitelio corneal es un proceso complejo que involucra la interacción de múltiples factores. Se ha observado que el epitelio corneal puede regenerarse de manera completa después de una lesión. Sin embargo, la cicatrización corneal puede ocurrir en casos de lesiones graves. La cicatrización corneal es un proceso inflamatorio que involucra la formación de un epitelio corneal persistente y la formación de una membrana basal.

La cicatrización corneal se caracteriza por la formación de una membrana basal persistente y la formación de una membrana basal persistente. La cicatrización corneal puede ocurrir en casos de lesiones graves, como en la cirugía refractiva con láser. La cicatrización corneal puede ser un problema en la recuperación del paciente y puede causar una disminución de la visión.

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the lack of mechanical influences of the upper eyelid that polishes the corneal surface with blinking. Epithelial hyperplasia in PRK is associated with deep stromal ablation depths and with small ablation zones (4.00–4.5 mm) because there is a marked curvature change in the edges of the ablated area. When ablation zones are large (6.00 mm), they have less demarcated contours, and thus, the change in epithelial thickness is minimal. Table 1 shows the variation of central corneal thickness with different surgery techniques published in the scientific literature. Erie and colleagues proved that, after PRK, the central epithelial thickness returned to preoperative levels at 1 month. However, it continued to progressively increase during the first year, being 21% thicker at that time. This result is similar to the 22% thickness increase seen in LASIK by Erie and colleagues. However, the time required for thickness stabilization differs between the two techniques, due to the complex interaction of epithelial cells and activated keratocytes in PRK. According to Patel et al., central corneal epithelium in LASIK increased 24% during the first year after surgery and remained stable during the next 7 years. In PRK, corneal thickness continued to increase at 1 month, 1 year and 7 years (442±39 μm, 464±44 μm, 471±45 μm; respectively). Recently, Ivarsen et al. have concluded that in PRK and LASIK, the epithelial thickness increases 15%–20% after surgery, but the epithelial changes in LASIK occur during the first week and remain unaltered during the following 3 years. It has been suggested that epithelial hyperplasia can induce a reduction of post-operative refractive effect. Erie showed myopic regression significantly associated with epithelial thickness increase. Nevertheless, Ivarsen et al. did not find any correlation between changes in epithelial thickness and changes in refraction after PRK or LASIK, probably because of the small size of their sample.

**Stromal Wound Healing Following PRK**

Stroma occupies approximately the 90% of corneal thickness, and it can be subdivided into three continuous layers: anterior, middle and posterior. The corneal stroma is built up from collagen fibers, ground substance, keratocytes and nerve fibers. Keratocytes – corneal stromal cells – play a major role in maintaining corneal transparency, and synthesizing the components of the extracellular matrix (ECM). Active keratocytes produce collagen and proteoglycans to form the ECM after stromal injury. The human stromal cornea contains collagen type I, V and VI. Type I is predominant (75%), followed by type VI (approximately, 17%). Type III collagen appears in inflammatory events or during wound healing. Proteoglycans participate in collagen fibrillogenesis and matrix assembly. After corneal injury, newly produced collagen fibers tend to have larger diameters, as they contain high levels of dermatan sulphate (a type of proteoglycan) that lasts up to 6 months.

Stromal keratocytes are normally quiescent or inactive, and are the second cells involved in the process of corneal regeneration, just after corneal epithelial cells. After PRK, keratocytes underlying the wound disappear by apoptosis due to a stress exposure. During the first 24 h after injury, macrophages, monocytes, T cells and polymorphonuclear cells infiltrate the area and remove damaged cells. Metalloproteinases (MMPs) and the plasminogen activator system remove the affected extracellular matrix. The MMPs are proteolytic enzymes secreted by active keratocytes or fibroblasts, and degrade complex molecules of the extracellular matrix. Although nine types of MMPs exist, in the cornea only four MMPs are important, being MMP-1 the most relevant. MMP-8 concentration has been observed to be significantly elevated in the second day after PRK (P= .001). The remaining keratocytes, adjacent to wound borders, are activated in response to various cytokines released by cells in upper layers, such as interleukin (IL)-1, and growth factors like tumor necrosis factor (TNF), fibroblast growth factor (FGF), platelet-derived growth factor (PDGF), epithelial growth factor (EGF), and transforming growth factor (TGF). These growth factors are normal components of the tear and corneal cells, produced by the lacrimal gland and regulate a variety of processes involved in homeostasis and corneal wound healing, including migration, mitosis and cell differentiation. Particularly, transformation growth factor beta (TGF-beta) seems to transform active keratocytes into myofibroblasts that appear at later stages of stromal heal-

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Figure 1 Corneal alterations and first steps of wound healing following PRK.
The density of keratocytes varies across the stroma. It is estimated that in the anterior stroma the density is 5%–10% greater than in middle and posterior stroma.\(^7\)\(^9\)\(^,\)\(^8\) It has been documented that a corneal stroma rich in keratocytes prevents the epithelial corneal infection or, at least, minimizes the extension of the infection.\(^2\)\(^,\)\(^5\)\(^,\)\(^10\) After PRK, the anterior keratocyte population drastically diminishes, and the distribution and shape is greatly altered.\(^2\)\(^,\)\(^10\)\(^,\)\(^34\)\(^,\)\(^40\)\(^,\)\(^42\)\(^,\)\(^81\)\(^,\)\(^82\) In confocal microscopy, high reflectance, hyperplasticity, hypertrophy and a decrease in the contrast of the anterior stromal keratocytes can be observed.\(^34\)\(^,\)\(^62\)\(^,\)\(^63\)\(^,\)\(^83\) Human histological studies confirm that the decrease of anterior stromal keratocytes in humans and animal respond similarly.\(^34\)\(^,\)\(^62\)\(^,\)\(^63\) Table 2 shows the variation of anterior, posterior and total keratocyte density in the different studies published in the scientific literature.\(^62\)\(^,\)\(^81\)\(^,\)\(^82\) Table 2 confirmed that after 5 years of PRK there were evidences of keratocyte density loss in middle and posterior stroma. They observed a reduction of 20%–24% in the posterior stroma \((P<.05)\) although they claimed that this loss was not completely evident. Keratocyte density in the anterior 10% of the stroma continues to decrease 5% per year between 1 and 3 years after PRK.\(^5\)\(^,\)\(^10\) Erie\(^1\) reported a progressive decline in anterior stromal keratocytes, becoming significant at 36 months after PRK \((P=.02)\). In contrast, middle and posterior keratocyte densities remained unchanged between 1 and 3 years after PRK.\(^5\)\(^,\)\(^10\) In another study, Erie\(^1\)\(^,\)\(^10\) proved that the keratocyte density in the anterior 10% of the stroma, decreased at 6, 12, 24 and 36 months \((41\%, 40\%, 43\%, 45\%;\) respectively) after PRK, compared to pre-PRK. In a posterior longer-term study, Erie\(^1\)\(^,\)\(^10\) demonstrated a similar decreasing pace in anterior keratocyte density: 40%, 42%, 45%, and 47% at 6 months, 2 years, 3 years, and 5 years \((P<.001)\). Amoozadeh et al.\(^40\) found a reduction in keratocyte density 6 months after surgery, but the loss was similar for LASIK and PRK interventions: in

<table>
<thead>
<tr>
<th>Study</th>
<th>Technique</th>
<th>1 month</th>
<th>3 months</th>
<th>6 months</th>
<th>1 year</th>
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<th>3 years</th>
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<td>529±31</td>
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Table 1: Variation of Central Corneal Thickness, Mean±SD or Range (μm).

PRK produces oxygen free radicals, secondary to the exposure of ultraviolet radiation, thermal increase, and polymorphonuclear cell infiltration.\(^7\)\(^,\)\(^10\) Free oxygen radicals may interact with lipid components, nucleoic acids, and sulphur contained in enzymes,\(^7\)\(^,\)\(^8\) and particularly with reactive oxygen species (ROE) that are considered to produce the most reactive and cytotoxic damage. In fact, they have been described as a partial cause of keratocyte apoptosis.\(^7\) Among the antioxidant enzymes that protect the cornea from radicals, superoxide dismutase (SOD), glutathione peroxidase (Gpx) and catalase are the most relevant.\(^7\)\(^,\)\(^8\) Ascorbic acid and \(\alpha\)-alpha-tocopherol (Vitamin E) also prevents from the effects of free radicals.\(^7\) Corneal epithelial ascorbic acid absorbs ultraviolet radiation, protecting keratocytes, but high or altered corneal ascorbate levels in the human cornea after PRK, may produce accelerated keratocyte death.\(^5\) In rabbit corneas, decreased activity of SOD and Gpx enzymes has been proved after refractive surgery.\(^10\) For this reason, additional antioxidant enzymes seem to be involved in reducing corneal oxidative stress following PRK. 1-cys peroxiredoxin (1-cys Prx) may be an important enzyme involved in the differentiation, migration and proliferation of epithelial cells. 1-cysPrx increases 4 h after PRK and remains in high levels until 7 days after PRK.\(^7\)

Myofibroblasts can be identified through the expression of \(\alpha\)-smooth muscle actin (SMA).\(^45\)
anterior stroma, 34.7% versus 31.13% (P>.05) and posterior stroma 0.31% versus 0.02% (P>.05), respectively. However, other studies have seen differences between PRK and LASIK, probably associated with the more superficial ablation in PRK.\(^5,25,40\) The consequences of keratocyte density loss after PRK are still unknown, but the visual acuity and corneal clarity seem to be preserved.\(^5\)

After the initial depletion of anterior stromal keratocytes, an increase in the keratocyte density is observed over time, probably secondary to mitosis, cellular migration, or reproduction of keratocytes and myofibroblasts.\(^5,9,10,34,51,86\) Following apoptotic keratocyte loss, the first morphological changes of remaining keratocytes that can be histologically observed, are an increase in cell size and an increase in the size and the number of nucleoli, rough endoplasmatic reticula, mitochondria, free ribosomes and Golgi complexes, indicating an active state.\(^65\) These keratocytes quickly repopulate the anterior stroma, and return to similar preoperative levels.\(^1,5,10,34\) Several studies using confocal microscopy have analyzed the keratocyte density after PRK. Corbett et al.\(^68\) found that at 2 days after PRK the anterior keratocyte density was increased 50%, 100% at 1 month, and returned to preoperative levels at 6 months. Frueh et al.\(^95\) concluded that the anterior keratocyte density increased 15% at 1 and 4 months after PRK, and returned to preoperative levels 1 year after PRK. Similarly, Erie et al. found an increase of 20% in the anterior stroma at 3 months after PRK.\(^2,87\) According to the results of Corbett et al.\(^68\) and Erie et al.,\(^88\) anterior keratocytes proliferation begins 1 month after PRK, with a pick at 3 months, and return to preoperative levels at 6 months.

### Corneal Haze

Corneal haze reduces corneal transparency at variable degrees.\(^90,91\) Subepithelial haze occurs in all patients 1 month after PRK, reaching the greatest intensity at 3–6 months, and gradually decreases from then on.\(^2,8,54,92\) Yet, some authors affirm that it begins to decrease at 12–24 months after PRK.\(^8,92\) Corneal haze is more common after correction of high myopia (>−6.00 D), and it is rarely seen after correction of <−6.00 D or <+4.00 D of hyperopia.\(^43,71,91\) Besides the ablation depth, the severity of corneal haze is correlated with excessive ocular UV-B radiation, duration of the epithelial defect, postoperative steroid treatment, male sex and with certain population with brown irises.\(^7,16,19,21,24,28,71,81,86,93–95\) PRK presents higher corneal haze incidence than LASIK, probably because of the destruction of the basement membrane.\(^8,43,93\) In the presence of damaged epithelial cells and basement membrane, cytokines and growth factors can easily flow from epithelium to anterior stroma.\(^45,96\) Cytokines released from epithelial cells activate keratocytes, as mentioned in a previous section, which synthesize large diameter collagen fibrils.\(^8,11,33,73,81\) Abnormally deposited extracellular matrix implies the development of corneal opacity.\(^59\) Moreover, active keratocytes present a high reflectance that also contributes to the decrease in corneal transparency. In addition, subepithelial vacuolation, deposit materials like proteoglycans, hyaluronic acid and collagen Type IV are involved in the formation of the corneal haze in advanced stages.\(^33,77\) Plasminogen activator–plasmin...
system degrades the damaged ECM, and extended low levels beyond the third day after PRK causes corneal haze formation. Guerriero et al.58 affirm that the loss of collagen type IV is related to the activation of keratocytes in vivo and in vitro, and Winkler et al.53 and Mohrenfels et al.58 emphasize on the role of type IV collagen in the development of corneal cloudiness. Secondary ultraviolet B (UV-B) exposure, originating from sun or solarium is a causal factor for aforementioned abnormal proteoglycan deposition and associated augmented corneal thickness.59

On the other hand, myofibroblast, derivatives of TGF-beta responding keratocytes, are thought to be the first biological event for corneal haze formation.51,93,94 Myofibroblasts play an essential role in the recovery of the corneal integrity after penetrating injury, mainly in advanced stages.22 They secrete extracellular matrix, contract wounds and have the ability to generate adhesion structures with the surrounding substrate.71 TGF-beta also induces the expression of connective tissue growth factor (CTGF), which mediates collagen synthesis, and along with myofibroblasts regulates the corneal wound healing, and may promote scar formation.100 After PRK, myofibroblasts appear as a pathological response to injury,71 and their decreased transparency roots in the low intracellular content of crystalline.102 Irregular surface has also been related to high incidence of corneal haze,94,102 and higher irregularity is seen with increasing dioptic corrections in PRK.103 Interestingly, surface irregularity is positively correlated with myofibroblast density in the anterior stroma.43 In normal corneal wound healing, complete regeneration of the basal membrane after PRK occurs within 6–8 weeks in rabbits,104 which limits the access of growth factors to the stroma29 and, consequently, myofibroblasts commit apoptosis modulated by IL-1.57 Therefore, the presence of myofibroblast, and subsequent corneal haze, is largely dependent upon the restoration of the basement membrane.43,103

Corneal haze has been traditionally measured in the slit-lamp, and graded with diverse scales, like Hanna’s scale. The new technology leads us to use automated instruments for corneal haze measurement. In vivo confocal microscopy is a reliable tool, as far as standardized methods are used.56 It is the most widely used objective method in clinical setting for haze measurement. In the last years, alternative techniques have come out. Confocal imaging of second harmonic-generated (SHG) signals has been shown to be sensitive in measuring corneal fibrosis after refractive surgery.105 Recently, the densiometry program of Pentacam Scheimpflug imaging system (Oculus Optikgeräte GmbH) has been proved to be a useful method for measuring corneal haze.108

Visual Disturbances of Corneal Haze

The corneal haze produces a reduction of low contrast visual acuity and night vision symptoms that, in the vast majority of situations, improve with time.53 It is possible to see corneal haze formation after PRK by means of confocal microscopy, observed as a decrease in the contrast of the image and an increase in reflectivity.81 Bohnke et al.81 using a Tandem scanning confocal microscopy, correlated corneal haze and anterior stromal reflectivity. However, the tandem scanning confocal microscopy is not able to detect acellular regions of the anterior stroma early after PRK when epithelium and sub-basal plexus are not formed.109 Although corneal haze in humans is less pronounced than in animal models, if corneal haze persists and affects significantly to the corneal transparency, it causes light scatter.94,109 For this reason, corneal haze may be described and analyzed through back light scattering (backscatter).81,110 It also causes irregular astigmatism,1,54,93 and subsequent loss of corrected distance visual acuity (CDVA).86

The regression of the refractive error may be produced by epithelial irregularity, alterations in the keratocyte density or subepithelial deposits. Myopic regression occurs in 78% of eyes in the first 12 months after PRK.2 Table 3 shows the mean spherical equivalent changes reported in different scientific studies. In the first week after PRK, epithelial irregularity causes a reduction in visual quality.88 During the first month, altered keratocytes decrease contrast sensitivity, mainly in high frequencies, and cause glare. During the next 2 months, subepithelial deposits produce a decrease in contrast sensitivity, especially in low frequencies.5,88 Ginis et al.4 reported that subepithelial deposits are the first factor that contributes to the development of corneal scatter. The visual quality is affected temporarily, although there is evidence that in some cases it persists for more than 1 year.43,97 In order to avoid a decrease in the visual quality, all postoperative efforts must go oriented to control the subepithelial matter.88 The corneal epithelium does not seem to contribute significantly to the refractive change after PRK, although some studies suggest that epithelial thickening may produce myopic regression,2 even 5 years after PRK.10 Moller-Pedersen et al.55 and Cua and Pepose52 suggested that new keratocytes growth in central cornea or postoperative corneal scarring is likely to be the main causes of myopic regression in ablations of 6 mm. In agreement with this hypothesis, Moller-Pedersen et al.55 demonstrated that hyperopic changes were the direct result of a stromal thinning. Erie4 found an increase of 12 μm of epithelial thickness at 12 months after PRK that was associated with a myopic regression of –0.41 diopters but no correlation was found between stromal thickening and myopic regression; however, the combined effect of epithelial and stromal thickening was correlated with myopic regression.

Regeneration of Corneal Innervation

The cornea is the most innervated tissue of the human body, and these sensory nerves are derived from the ophthalmic branch of the trigeminal nerve fibers.4,111 Corneal sensory nerves penetrate the limbus and form nerve bundles in the anterior third of the stroma. Once there, they run perpendicularly to cross Bowman’s membrane, and form the sub-basal nerve plexus as a network between the basal epithelial cells and Bowman’s layer (Fig. 2).49,111 Corneal nerve fibers, if visualized using confocal microscopy in normal conditions, show high reflectivity across the corneal stroma with a rectilinear pattern. Subepithelial nerve fibers, on the other hand, are thinner than stromal nerve fibers. Corneal fibers are considered primarily nociceptive (70%), followed by mechanosensitive fibers (20%).112 In PRK, phot-
### Table 3  Mean Spherical Equivalent After Surgery, Mean±SD or Range (Diopters).

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<th>Technique</th>
<th>Preop</th>
<th>1 month</th>
<th>6 months</th>
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<td>−0.28±0.91</td>
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<td>(−0.50 to 1.50)</td>
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<td>PRK+MMCa</td>
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<td>−0.28±0.61</td>
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<td>Ghirlando et al. (2007)</td>
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<td>−3.95±1.29 (−1.95 to −6.40)</td>
<td>+0.22±0.79</td>
<td>(−0.50 to 2.88)</td>
<td>−0.17±0.35</td>
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<td>PRK+MMCb</td>
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<td>−0.08±0.38</td>
<td>(−0.75 to −0.75)</td>
<td>−0.18±0.35</td>
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<tr>
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<td>LASIK</td>
<td>−6.5±2.5 (−11.0 to −2.0)</td>
<td>−0.1±0.5</td>
<td>(−1.0 to +1.0)</td>
<td>−0.2±0.5</td>
<td>(−1.25 to +0.75)</td>
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<td>(−1.0 to +0.75)</td>
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<td>PRK</td>
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<td>(−0.5 to +1.0)</td>
<td>−0.3±0.3</td>
<td>(−0.87 to plano)</td>
<td>−0.4±0.4</td>
<td>(−1.25 to plano)</td>
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<td>(−1.25 to +0.75)</td>
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<td>−3.7±1.4 (−5.75 to −1.25)</td>
<td>−0.1±0.3</td>
<td>(−0.5 to +1.0)</td>
<td>−0.3±0.3</td>
<td>(−0.87 to plano)</td>
<td>−0.4±0.4</td>
<td>(−1.25 to plano)</td>
<td>−0.3±0.2</td>
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<td>Lee et al. (2005)</td>
<td>PRK</td>
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<td>−0.46±1.01</td>
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<td>tPRK</td>
<td>−5.11±1.51 (−1.87 to −9.50)</td>
<td>0.59±0.78</td>
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<td>0.18±0.91</td>
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PRK, photorefractive keratectomy; MD, mechanical debridement; AAD, alcohol-assisted debridement; LASIK, laser in situ keratomileusis; MMC, mitomycin C; LASEK, laser-assisted subepithelial keratectomy; tPRK, trans-PRK.

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a 0.002%, 1 min.
b 0.02%, 2 min.
toablination severs nerves of the subbasal plexus and anterior stroma.\textsuperscript{2,83} It has been suggested that axotomy of corneal nerves might cause the decrease in keratocyte density after PRK.\textsuperscript{113,114} Corneal nerves directly innervate keratocytes and provide trophic support in normal conditions.\textsuperscript{115} Animal studies have proved that the regeneration of the corneal nerves after PRK occurs as a biphasic process. In the first stage, a subbasal plexus originates from the cut end of subepithelial plexus, and the fine neurites run centrally with migrating cells.\textsuperscript{116,117} In the next phase, this transient plexus degenerates, and stromal originated nerves take place.\textsuperscript{117} Subbasal nervous plexus can have a significant influence on the regulation of epithelial healing.\textsuperscript{14} Substances like chemokines, proteases and neuropeptides are released after corneal injury.\textsuperscript{8,24,25,50,51,118} and it is postulated that neuropeptides like substance P (SP) and calcitonin-gene related peptide (CGRP) contribute to corneal wound healing.\textsuperscript{112} Corneal nerves also influence the production of collagen type VII, necessary for the anchoring of the epithelium to the stroma.\textsuperscript{119} Conversely, injured epithelial cells release nerve growth factor (NGF) that stimulates nerve regeneration.

Approximately, at 8 weeks after PRK, sub-epithelial nerve fibers are visible on the edges. Erie,\textsuperscript{2} using tandem scanning confocal microscope, visualized subbasal nerve fiber bundles in 17\% of the corneas at 1 month after PRK. However, he noted that the density of these nerve fibers was 98\% less than preoperatively.\textsuperscript{2} After about 3 months of the surgery, no branched nerve fibers can be visualized in the center of the zone of ablation. Changes in subepithelial plexus and stromal trunks begin to appear 2--4 months postoperatively.\textsuperscript{8} At 6--8 months after the intervention of PRK subepithelial nerve regeneration is almost complete.\textsuperscript{8,111,120} although changes in the structure of the corneal nerves can be appreciated by confocal microscopy up to 12 months postoperatively.\textsuperscript{120} However, nerve density continues to improve until 12 months after surgery, and returns to the preoperative values at 2 years.\textsuperscript{121} According to Moilanen et al.,\textsuperscript{122} in 71\% of cases the central branch ing postoperatively was comparable to control subjects at 5 years (P=.56). Erie\textsuperscript{2} proved that subbasal nerve density was reduced at 3, 6 and 12 months (87\%, 75\%, 60\%, respectively) after PRK, and returned to preoperative levels at 24 and 36 months postoperatively. Subsequently, Erie et al.\textsuperscript{83} in a prospective 5-year longitudinal clinical trial, proved with confocal microscopy that the recovery of subbasal nerve density in central cornea was faster in PRK than in LASIK. The authors observed that subbasal corneal density was reduced by 59\% at 1 year after PRK (2764±1321 $\mu$m/mm$^2$) compared to preoperatively (6786±1948 $\mu$m/mm$^2$; P<.001). Sub-basal nervous plexus was almost recovered 2 years after PRK (6242±1763 $\mu$m/mm$^2$), and remained unchanged at 3 years (6358±2447 $\mu$m/mm$^2$) and 5 years (5903±3086 $\mu$m/mm$^2$).\textsuperscript{83} In LASIK, they observed that subbasal corneal density was reduced by 34\% at 3 years (P<.001),\textsuperscript{83} with values at 5 years postoperatively comparable to those obtained preoperatively (5903±3086 $\mu$m/mm$^2$).\textsuperscript{83} It is worth to note that in this study, the corneal flap was created using a mechanical microkeratome.\textsuperscript{83} As the new technology allows making corneal flaps with laser instead of with a mechanical microkeratome, it is possible that studies in the near future report a faster corneal nerve recovery after LASIK.

When the process of corneal nerve regeneration finalizes, morphological abnormalities are often observed.\textsuperscript{8,83,122} According to Erie,\textsuperscript{2} in the first 6 months after PRK the central subbasal nerves are organized in horizontal or oblique orientation. However, between 6 and 12 months, the subbasal nerve orientationforums and comes to vertical orientation. In dry eye conditions, Esquenazi et al.\textsuperscript{25} observed active keratocytes, and they expressed nerve growth factor (NGF). NGF stimulates the proliferation of basal epithelial cells in normal conditions. Active keratocytes provoke an overexpression of NGF, which leads to abnormal findings in corneal nerves, such as hypertrophy.\textsuperscript{27} They also found higher nerve tortuosity, higher number of nerve beards, and the presence of nerve sprouts in desiccating environment group,\textsuperscript{22} which

![Figure 2](https://example.com/figure2.png)  
**Figure 2** Schematic representation of corneal stromal nerves and subbasal plexus in human cornea. (A) Frontal view. (B) Cross section.
Corneal Pain and Sensitivity

Photoblation severs corneal nerves, disrupting the lacrimal functional unit (LFU). LFU is constituted by the lachrymal gland, ocular surface and innervation. It regulates tear secretion, and affects its composition.123 Thereby, photoblation produces transitory dry eye, deterioration of corneal barrier function and alteration in corneal sensitivity.83,111,124 A reduction of the tear flow after PRK has been proved using Schirmer test.14 According to Erie et al.,4 LASIK presents higher prevalence of postoperative dry eye, altered corneal epithelium and tear film than PRK. Dry eye has been associated with low corneal sensitivity.123,126 Different devices are available to measure corneal sensitivity, as Cochet-Bonnet esthesiometry or non-contact gas esthesiometry. Cochet-Bonnet esthesiometry only stimulates mechanosensory fibers, whereas non-contact gas esthesiometry measures activation thresholds of nociceptors using controlled chemical, thermal and mechanical pulses. Non-contact gas esthesiometry is, therefore, a more sensitive device for measuring alterations in corneal sensitivity. Still, Cochet-Bonnet esthesiometry is more widely used, and controversy remains about the time course of the corneal sensitivity recovery after PRK with this device. Kauffmann et al.130 affirm that the recovery of corneal sensitivity usually starts at 4–6 weeks, completing approximately within 6–12 months following PRK. However, Erie et al.4 claim that the recovery of corneal sensitivity is completed from 3 months to 1 year after PRK. Hypoesthesia is often expected until 3 months after surgery, due to the loss of corneal nerves.8 On the other hand, Gällar et al.127 measured corneal mechanical and chemical sensitivity following PRK with non-contact gas esthesiometer, and found that both types of sensitivities were reduced even 5 years postoperatively, achieving normal values in 10 years. Despite the diminished corneal sensitivity, intense pain is usually present hours after PRK.129 Gällar et al.129 attributed corneal pain and discomfort sensations to the altered functionality of corneal nerves. They recorded spontaneous activity and modified responsiveness in corneal fibers of cats that underwent PRK.129 Experimental evidences support the idea that ongoing activity evokes spontaneous pain sensations.130,131

Epithelial Removal Techniques

In PRK, previous to the impact of laser energy over the cornea, the corneal epithelium has to be removed. The removal of the corneal epithelium is carried out mainly with epithelial mechanical scraping using chemical agents like diluted ethanol solution,9 through a rotary brush or using the laser itself – known as transepithelial ablation (Fig. 3) 16,21,22,29,63,128,132,133 The epithelial scraping has postoperative adverse effects like pain, myopic regression or corneal haze. Some modification in PRK technique can alter the wound healing response with the aim of minimizing the adverse effects.23 The exposition to agents such as ethanol can produce an increase in the inflammatory response and more damage to the anterior stromal keratocytes that could increase the haze formation.21,24 Yet, controversy remains in the scientific literature because other authors affirm that alcohol-assisted epithelial removal produces less inflammation, favoring epithelial regeneration and preventing corneal haze or keratoacyte apoptosis.9,63 Esquenazi et al.21 proved that the epithelial scraping might be associated with an increase in the number of reflective structures in the stroma, mainly in corneas with ocular dryness after PRK. The laser-scrape epithelial removal decreases the degree of keratocyte apoptosis, producing a less pronounced loss of superficial keratocytes.21 However, the irrigation with cold balanced salt solution (BSS) may alter the keratocyte apoptosis in the retroablation zone. The time necessary for mechanical debridement is greater than the time required for laser or alcohol scrape techniques, even for expert surgeons.18 Mechanical debridement is related to stromal dehydration and disappearance of anterior stromal keratocytes.6,63 This loss provokes an increase of cells in the underlying stroma, causing stromal hyperplasia and haze formation.134 Einollahi et al.63 found faster mean epithelial healing time in the alcohol-assisted group than in the mechanical group (3.0±0.3 versus 3.2±0.4 days, P<0.001). They observed greater anterior retroablation stromal keratocyte density in the mechanical group than in the alcohol-assisted groups at 3 months (704.3±119.9 cells/mm² versus 743.3±103.7 cells/mm², P<0.05) and at 6 months (643.8±134.4 cells/mm² versus 696.7±129.6 cells/mm², P=0.02).15 In the same study, Bahram et al. did not found statistically significant differences in middle and posterior keratocyte density between the mechanical and alcohol-assisted groups.63 They also proved that mechanical and alcohol-assisted epithelial debridement after PRK present similar visual and refractive outcomes in patients with mild myopia,63 in agreement with the results of Goreishi et al.135 They reported similar safety and efficacy with alcohol-assisted and mechanical debridement in a 1250 eye sample, but anterior keratocyte density was not assessed in this study.136

Laser-assisted subepithelial keratomileusis (LASEK) was developed in order to reduce corneal pain and haze formation associated with PRK, and to accelerate visual recovery. Epithelial delamination with diluted alcohol showed in an electronic microscope study that was able to leave a smooth surface, ideal for LASEK intervention.136 It seems that a regular surface before laser application helps corneal healing and prevents haze.137 Chen et al.138 contrasted these findings in a later study, and showed a high variability
in morphological changes after diluted alcohol treatment, dependent upon concentration and time. Cell viability was affected when alcohol exceeded its concentration by 25% or 25-s exposure. They have been conducted in vitro, and the complex interactions of tear film and corneal surface were not considered. In vivo studies do not show any difference between LASEK and PRK. Lee et al. evaluated epithelial healing, postoperative pain and visual outcomes using epithelial mechanical (conventional PRK), transepithelial PRK and 20% diluted alcohol laser-assisted subepithelial keratomeleusis (LASEK) with flap repositioning. After 6 months, they found little differences in clinical outcomes between the 3 techniques, noting a slight overcorrection in the transepithelial PRK and slight undercorrection in LASEK. Corneal pain and subepithelial haze results were similar. Subsequently, Ghanem et al. proved in a prospective randomized double-masked study that the reepithelization was faster in a PRK group compared with a butterfly LASEK group, even though epithelial semi-discs were repositioned intraoperatively in LASEK group. They also found lower pain level in PRK group, but pain scores and ocular discomfort were not statistically different from butterfly LASEK (3.31±4.09 versus. 4.43±4.27; P=.18). It has been proven in animal studies that transepithelial ablation produces a uniform surface for corneal regeneration, and prevents keratocyte apoptosis, reducing the risk of corneal haze. Wang et al. presented promising preliminary results of SCHWIND-ESIRIS excimer laser for transepithelial ablation, but the flawed design of the study makes difficult to assess the real value of this technique. Later, Aslanides et al. proved in humans that transepithelial ablation was safer than the epithelial mechanical scraping using chemical agents as alcohol, as it provides a faster epithelial healing, less postoperative pain and less corneal haze at 1 week (P=.07), and at 1, 3, and 6 months after surgery (P<.05). In addition, they observed an improvement of 3 Snellen lines in visual acuity on day 3 in the modified transepithelial PRK (all-surface laser ablation) group compared to conventional alcohol-assisted PRK group (0.4 versus 0.2; P<.05). Transepithelial ablation also resulted in better corrected distance visual acuity (DCVA) than conventional alcohol-assisted PRK (33% versus 13%, respectively, P<.05), although differences in higher order aberrations were not statistically significant.

Amniotic Membrane Transplantation

Apart from the above mentioned techniques, amniotic membrane transplantation reduces the inflammation after PRK, prevents polymorphonuclear cell infiltration, produces less peroxidation, avoids keratocyte apoptosis and stimulates corneal epithelialization. It is usually combined with PRK to treat corneal dystrophies, corneal degenerations, scars, keratopathsies, or even to treat corneal haze secondary to PRK. The amniotic membrane restricts the influx of polymorphonuclear cells (PMCs) to the patch. PMCs adhere to the amniotic membrane and eventually commit apoptosis. This is a physiological way of suppressing corneal inflammation. In addition, amniotic membrane has intrinsic keratocyte growth factors, EGF and neurotrophins that promote epithelization. It also suppresses TGF-beta1, collagen III and fibronectin. Taken together, amniotic membrane has a potent anti-scarring effect that reduces corneal haze formation, as demonstrated in animal studies.
Agents to Enhance Wound Healing

The wound healing response may be altered by the prophylactic application of a topical solution of mitomycin-C (MMC) immediately after the laser ablation, in order to avoid or minimize myofibroblast activation. MMC is an antineoplastic antibiotic agent of the family of anti-tumor quinolones and derived from Streptomyces caespitosus. It is a potent DNA crosslinker: it inhibits the replication of deoxyribonucleic acid (DNA).

MMC is applied for a short duration (0.2% MMC) for 5 min at the end of the procedure. This procedure is applied to the entire 3 groups of human corneas: without MMC, with MMC (0.2 mg/ml) for 1 min, and with MMC (0.2 mg/ml) for 2 min. The 2 min MMC group had a thinner epithelium than the 1 min group and the MMC application group (P < 0.0001).

Rajan et al. analyzed the effects of MMC after corneal injury in 9 human corneas. The MMC group had a thinner epithelium and a better corneal healing response than the control group.

Shojaei et al. observed a delay in keratocyte regeneration after MMC application (P < 0.005). Midena et al. proved by means of confocal microscopy that the application of 0.02% MMC produced a considerable decrease in anterior stromal keratocytes, but there is no evidence of this decline in the posterior stromal keratocytes. Subsequently, Thornton et al. observed a keratocyte loss in the anterior stroma 1 month and 6 months after PRK with standard MMC concentrations (0.02%). Razmjoo et al. did not find significant reduction in keratocyte density after application of 0.02% mitomycin C (MMC). The dose of MMC is associated with the grade of refractive error. Thornton et al. believe that for high myopia corrections (> −6.00 D) standard concentration of topical MMC (0.02%) may be used, whereas for moderate myopia (−3.00 to −5.90 D) low dose of MMC (0.002%) may be considered, although it seems that intermediate dose of MMC (0.02%) is more effective than 0.002% for moderate myopia.

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The cytotoxicity of MMC increases with cumulative doses, and when MMC is combined with ethanol, which increments the apoptosis of keratocytes. Few complications have been associated with the use of MMC. There is an exception to the decrease in the short term of the keratocyte density. However, some complications have been documented at the time of instillation or after some weeks. Although unusual, scleral ulceration, non-healing conjunctivae and complications associated with high MMC doses (0.04%) or prolonged postoperative topical use may appear, because high doses of MMC suppress cellular RNA replication and protein synthesis.

As MMC is applied in the stromal bed, it seems that it might penetrate into the anterior chamber, because cytotoxic effects on the ciliary body epithelium have been reported.

The endothelium is the inner layer of the cornea. Endothelial cells have a hexagonal or polygonal shape, and they are homogeneously distributed, without signs of polymegatasm and pleomorphism in normal conditions. Endothelial cells are not able to regenerate, and a reduction in the number of cells is seen with age. After PRK, endothelial structure, shape and density remain unaltered. Table 4 shows the variation of endothelial cells in the different studies published in the scientific literature. Polymegatasm or pleomorphism, if present, may be secondary to still unknown corneal metabolism.

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New generation quinolones, instead of preventing corneal haze, are used as prophylactic antibiotics to avoid corneal infections after refractive surgery. 72-74 They also enhance the rate of corneal recovery. Fourth generation fluoroquinolones like gatifloxacin (Zymar, Allergan, Irvine, California) and moxifloxacin (Vigamox, Alcon Laboratories, Fort Worth, Texas) have been demonstrated to mediate faster corneal healing, 72 without evident differences between both of them in terms of visual outcomes. 118

### Bandage Contact Lenses

After PRK, the corneal surface needs between 2 and 4 days to regenerate, 7 and the vision may fluctuate for several weeks to months. If epithelial regeneration delays, the subepithelial haze increases; for this reason, an appropriate corneal reepithelization is crucial. 5,109 Reepithelization is the first step during corneal regeneration after PRK. 51 If the reepithelialization is facilitated with the appropriate contact lenses, visual acuity improves. 30,173 Although therapeutic contact lenses have been used for more than 40 years, PRK has increased their popularity. 23,30 One of the major disadvantages of PRK is the pain and discomfort during 1–3 days after intervention. 7,76,174 To ease off the postoperative pain and discomfort, and to promote epithelial healing, bandage contact lenses are fitted for 3–5 days after surgery. 12,23,30,11 Other techniques and medications has been proposed in order to reduce corneal pain like occlusive pressure patching, but the bandage contact lenses are still the gold standard. 173 Bandage contact lenses are used to protect the epithelium from the eyelid, to reduce the haze formation, 11,173 to enhance epithelial healing, to control the sensation of pain, and to prevent epithelial erosions. 12,23,30,118 Faster reepithelialization produces a reduction of discomfort, facilitates visual recovery, and restores the corneal barrier to prevent infections. 12.

Because of the prolonged use of therapeutic contact lenses, and to assure the proper corneal metabolism, a high oxygen permeability (Dk/t) contact lens are used. 12,30,118,173 Silicone hydrogel contact lenses have a Dk/t coefficient 5–10-fold greater than conventional hydrogel lenses. 12 For this reason, silicone hydrogel bandage contact lenses are widely fitted, 7,12,30,31 and are the ones approved by the FDA for prolonged use after PRK. Currently, a variety of contact lenses are used as therapeutic soft contact lenses after PRK like Lotrafilcon A (Focus Night & Day, Ciba Vision), Lotrafilcon B (O 2 Optix, Ciba Vision), Senofilcon A (Acuvue Oasys, Vistakon Inc.), Balafilcon A, Omafilcon A (Proclear, Cooper Vision) and Senofilcon A. 12,23,30,31 Lotrafilcon B is approved by FDA for 6 days of continuous wear and Senofilcon A for 1 week of continuous wear, while Lotrafilcon A

### Table 4 Variation of Endothelial cell density After Surgery, Mean±SD or Range (cell/mm²).

<table>
<thead>
<tr>
<th>Study</th>
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<th>3 months</th>
<th>6 months</th>
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<td>Einollahi et al. (2011) 63</td>
<td>PRK with BSS</td>
<td>2819±203</td>
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<td>Wallau and Campos (2008) 159</td>
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<td>PRK with MMC</td>
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<td>Nassaralla et al. (2007) 28</td>
<td>Epi-LASIK</td>
<td>2769±158</td>
<td>2727±179</td>
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<td>Morales et al. (2006) 158</td>
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<td>2779±492</td>
<td>2711±555</td>
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PRK, photorefractive keratotomy; MMC, mitomycin C; BSS, balanced saline solution; MD, mechanical debridement; AAD, alcohol-assisted debridement; LASIK, laser in situ keratomileusis; Epi-LASIK, epipolis in situ keratomileusis.

* After radial keratotomy.
  b 0.002%, 1 min.
  c 0.02%, 2 min.
  d 0.02%, 30s.
  e 0.02%, 15s.
is approved for 30 days of continuous wear and therapeutic use.\textsuperscript{2,8,21} The therapeutic efficacy of the Lotraficion A after PRK has been intensively studied,\textsuperscript{12,23,30,31} and reduction of discomfort and faster corneal reepithelialization in 48 h have been described.\textsuperscript{12,23} Edwards et al.\textsuperscript{31} proved that Lotraficion A showed better best spectacle-correction visual acuity (BSCVA) than Omafliclon A, without statistically significant differences in contrast sensitivity or uncorrected visual acuity (UVA). Omafliclon A reduced the BSCVA in 40.4% of patients at 1 month, whereas Lotraficion A reduced the BSCVA in 18.6% of the patients ($P=.002$). The corneal pain was greater with Omafliclon A than with Lotraficion A at 1 day ($P=.000$) and 4 days postoperatively ($P=.027$).\textsuperscript{31} In contrast, an increase in corneal infiltrates with Lotraficion A was observed compared to Omafliclon A, and there was not a statistically significant difference in reepithelialization.\textsuperscript{31}

The authors suggested that corneal infiltrates might be a consequence of Lotraficion A’s rigidity due to its reduced water content (24%) versus 59% of Omafliclon A.\textsuperscript{31} Subsequently, Razmjoo et al.\textsuperscript{30} in a comparative study, found that the 50.3% of the eyes with Senoficlon A and 41.7% of the eyes with Lotraficion A completed the reepithelialization at day 5 ($P<.05$). Although there were not statistically significant differences in the rate of corneal reepithelialization between both contact lenses ($P>.05$), and the postoperative pain and discomfort index was significantly lower in Senoficlon A group ($P<.05$).\textsuperscript{30} They also compared the visual acuity between Senoficlon A and Lotraficion A after PRK, and proved that in both groups the UCVA was worse at 3 days than at day 1. However, the UCVA improved at day 5, with 97.7% reaching UCVA of 20/40. A feasible explanation is that, at day 3, the epithelial healing process is located in the center of the cornea.\textsuperscript{30} As only 44 patients were included, in future studies a larger size sample would be recommendable.

Bandage contact lenses also minimize corneal haze. Edwards et al.\textsuperscript{31} showed a minimum tendency to a high level of corneal haze with Omafliclon A compared with Lotraficion A ($P=.064$). However, all efforts are made to minimize the corneal haze intraoperatively, using cold balanced saline (BSS) and MMC. Application of BSS in the stromal body reduces the corneal pain and corneal haze;\textsuperscript{128} yet, the application of mitomycin-C (MMC) is more widely used.

Although bandage contact lenses have various advantages, the presence of silicone may produce irritation, increased protein and lipid deposits, and reduced wettability because of its hydrophobicity.\textsuperscript{31} A plasma treatment is given to enhance the hydrophilicity of Lotraficion A surface, but this technique is not completely effective.\textsuperscript{31} Bacterial keratitis and subepithelial infiltrates have been described with bandage contact lenses after PRK.\textsuperscript{12} The risk of infectious keratitis of soft contact lenses fitted for approximately 3 days is low, and antibiotics are prescribed to further minimize the risk.\textsuperscript{172}

**Corticosteroids and Non-steroidal Anti-inflammatory Agents (NSAIDs) Therapy**

It is necessary to distinguish between corneal haze that appears in the first weeks or months after PRK and pathological corneal haze that appears as a result of myofibroblasts.\textsuperscript{71} If the corneal haze persists over time, it may cause a corneal opacity and the thickening of the tissue that would result in a regression of the refractive error, decreased visual acuity and irregular astigmatism.\textsuperscript{13,34,67,71} Clinically significant corneal haze occurs in 0.5%–5% of the cases.\textsuperscript{109} Corneal haze that most commonly occurs after PRK is not clinically significant, and is not attributed to myofibroblasts.\textsuperscript{71,121} According to Wilson,\textsuperscript{71} in human corneas that develop late corneal haze after PRK, the resolution of the opacity is slow, and the restoration of the refractive correction is produced between 1 and 3 years postoperatively. It has been postulated that the extinction of corneal haze can be influenced by the disappearance of myofibroblasts, reabsorption of abnormal extracellular matrix (ECM) and restoration of normal corneal structure.\textsuperscript{71}

After surgery, a variety of drugs are prescribed to avoid corneal haze, for instance, corticosteroids – antiinflammatory to avoid the pain and inflammation-, plasmin inhibitors, growth factors or antimetabolites.\textsuperscript{7,115} Topical therapy after PRK prevents complications like keratitis, infections or corneal haze.\textsuperscript{177} The most common treatment after PRK to avoid the corneal inflammation is the application of corticosteroids.\textsuperscript{109,158} Corticosteroids are not recommended for long periods because of their side effects, like intraocular pressure (IOP) rise and the risk of cataracts.\textsuperscript{109,176,179} Javadi et al.\textsuperscript{180} reported a rise in the IOP using 0.1% betamethasone at 2 weeks post-PRK in a minority of patients. Furthermore, corticosteroids delay epithelial healing.\textsuperscript{179} When corneal haze appears 2–3 months after PRK, the clinical observations confirm that haze is “corticosteroid-responsive” in 10%–15% of patients.\textsuperscript{71} Researchers disagree about the benefit of corticosteroids to reduce the corneal haze after PRK.\textsuperscript{5,177} According to Wilson,\textsuperscript{71} the topical administration of 1% prednisolone acetate (Pred Forte) quickly removes the corneal opacity and produces a change in refractive error. In the remaining 85% or 90% of cases, the corticosteroids do not exert any change.\textsuperscript{71} Corticosteroids could be replaced by non-steroidal anti-inflammatory agents (NSAIDs), tranilast, cycloheximide or antioxidants like Vitamin E.\textsuperscript{109,177} NSAIDs are effective in reducing corneal pain, postoperative photophobia and inflammation.\textsuperscript{7,118} The inflammatory response is mediated by prostaglandins synthesized from arachidonic acid by cyclooxygenase 1 (COX-1) or cyclooxygenase 2 (COX-2).\textsuperscript{125} The anti-inflammatory and analgesic properties of the nonsteroidal anti-inflammatory drugs (NSAIDs) are achieved by the inhibition of COXs activity.\textsuperscript{7,118}

The use of certain steroidal and non-steroidal anti-inflammatory drugs (NSAID) delay reepithelialization and increase the risk of haze formation, although the results are still contradictory. Vetrugno et al.\textsuperscript{177} proved that 0.1% flurometholone acetate administered in the first day after PRK reduced corneal haze and myopic regression, particularly in high myopic patients. NSAIDs like diclofenac and ketorolac have shown reduction in the pain sensation,\textsuperscript{7,118} but also a significant delay in corneal reepithelialization after PRK.\textsuperscript{181} Nepafenac (Nevanac; Alcon Laboratories Inc., Ft Worth, Tex) is a new topical NSAID with greater corneal permeability that has been approved for the treatment of inflammation after surgery.\textsuperscript{7,181} Jalali et al.\textsuperscript{13} found that 0.1% Nepafenac did not increase haze formation, neither hamper corneal epithelial healing, but they did...
not found statistically significant differences in corneal reepithelization between nepafenac and non-nepafenac groups (P=.61). Caldwell et al.,18 in a randomized double-masked study, demonstrated that 0.1% nepafenac was safe for corneal reepithelialization, and reduced the post-operative pain at day 1 (0.76 versus 1.68) and day 2 (1.26 versus 2.23) compared with the placebo group (P<.0005). Other NSAIDs for corneal pain reduction are also available, like Bromfenac, Flurbiprofen sodium and Indomethacin.7

Despite the presence of complications is low, non-steroidal anti-inflammatory drugs (NSAIDs) may produce conjunctival hyperemia, transient burning, stinging, superficial punctate keratitis, epithelial defects, subepithelial infiltrates, corneal melting and perforation.7,17 However, Caldwell et al.,18 in a randomized double-masked study, proved that 0.1% nepafenac did not had adverse effects. Postoperative oral analgesics, like NSAIDs, are able to produce gastrointestinal, cardiovascular, respiratory and central nervous system complications.17,18 Another treatment that is widespread for the inhibition of inflammation and for treatment of dry eye is the Cyclosporine A, with doses from 0.05% to 2.00%.10 Nien et al.10 used Cyclosporine A 0.05% and prednisolone acetate 0.1% to compare the effect in corneal haze prevention in rabbit corneas following PRK. They concluded that Cyclosporine A did not have any effect, whereas prednisolone acetate was effective in reducing short-term corneal haze, but did not prevent corneal fibrosis.10

**Alternative Therapies for Corneal Haze Prevention**

The use of drugs does not completely suppress corneal haze formation after PRK. Research has focused on new therapies that could prevent corneal haze, like genetic evaluation of type IV collagens synthesis.25 Lumican and keratan genes have also been evaluated for management of subepithelial persistent corneal haze after PRK, but without a consistent finding.182 It has been postulated that vitamin E, probucol or heparin may inhibit collagen type IV synthesis, but they have not been approved for topical use because of their adverse effects.15 Vitamin E and amino acids play an important role in corneal reepithelialization and in the prevention of corneal haze and keratocyte apoptosis, especially in high myopia.10 A preliminary clinical trial concluded that oral supplementation with vitamin A and vitamin E accelerated the reepithelialization, and reduced corneal haze formation,193 but it seems that the topical administration of vitamin A alone do not have any effect.184 Alternative treatments to MMC that prevent corneal haze formation, but reduce less damage to keratocyte are bevacizumab and rapamycin.185 Subconjunctival injection of PRM-151 could presumably prevent corneal haze, as it inhibits the pro-fibrotic myofibroblast differentiation.186 Trichostatin A, similarly, prevents myofibroblast formation by inhibiting TGF-beta1.187 As cytokines and growth factors control the synthesis of collagen type IV, they might be also useful treatments for corneal haze prevention.25 PRK increases the release of leukocytes, TGF-β1, TNF-α and PDGF-BB in human tears during the first days of wound healing.18,30 TGF is a cytokine released by the lacrimal gland, corneal epithelium and conjunctival cells.179 Three forms of TGF-β exist (TGF-β1, TGF-β2 and TGF-β3) and each one is involved in the wound healing process in a different way. TGF-β1 is increased in early epithelial healing, and exerts an influence in the subepithelial fibrosis formation and activation of keratocytes after PRK.25 Bühren et al.199 proved that the application of anti-TGF-β in felines reduced the differentiation in vitro of keratocytes into myofibroblast, and corneal haze diminished. They suggested that this reduction in differentiation improved optical quality. The combination of the nerve growth factor (NGF) and decosahexaenoic acid stimulated the regeneration of basal epithelial cells in rabbits after PRK,20 which is imperative for a proper wound healing. Medduri et al.20 studied the effect of basic fibroblast growth factor (b-FGF) in a model of delayed healing after PRK.25 50 patients were enrolled in b-FGF eye drop treatment group and 50 patients in saline drops (placebo) group. They observed greater corneal epithelial healing in the b-FGF group than in the placebo group at 4 days (98% versus 72%, respectively) and 5 days after surgery (100% versus 92%, respectively).25

Artificial tears are the most widely used solution for corneal lubrication. However, they do not have biological components that promote corneal regeneration. In fact, they contain stabilizers, preservatives, or other additives that may induce toxic or allergic reactions.23 Blood derivatives as plasma rich in growth factors are an alternative to artificial tears, and have not possibility of rejection.26 Anitua et al.51 proved that plasma rich in growth factors obtained from patient’s blood enhanced corneal healing, and reduced the formation of corneal haze. The difference between the plasma rich in growth factors group (PRGF-Endoret) and control group was negligible at day 3. They attributed it to the increase of proliferative cells (Ki-67%) in the control group. They suggested that the increase in proliferative cells could be associated with epithelial hyperplasia observed at day 3 and 7 after PRK in control group.51 They also found that the epithelium of the PRGF-Endoret group was formed by 5–6 layers.51

**Corneal Nerve Regeneration and Neuropathic Corneal Pain Management**

The regeneration of the corneal nerves after PRK is associated with the improvement of cellular integrity.22 To date, few therapeutic treatments have been developed for nerve regeneration. Javaloy et al.108 investigated the benefits of topical platelet-rich plasma, but subbasal nerve density did not improve after 3 months of treatment compared to controls (P=.66). Studies in animal models have demonstrated more encouraging results. Esquenazi et al.21 studied the outcomes of the combination of nerve growth factor (NGF) and docosahexaenoic acid (DHA) in rabbits in promoting corneal nerve regeneration. They observed that this combination increased corneal nerve regeneration, as well as epithelial proliferation and decreased rose bengal staining compared to the application of NGF or DHA alone. Cortina et al.180 showed similar results with pigment epithelial-derived factor (PEDF) plus docosahexaenoic acid (DHA).
Moreover, this combination proved to enhance corneal sensitivity. Recent evidence suggests that peripheral nervous system regeneration and inflammatory processes share common pathways, and some degree of inflammation is required for neuroregeneration. Therefore, cyclosporine A and corticosteroid treatments could interfere in a proper nervous recovery.

When corneal nerve regeneration process fails, corneal neuropathic pain might take place. The up- and down-regulation of ion channels in axotomized nerves can change the excitability of fibers, and produce spontaneous discharges and altered sensibility to exogenous stimuli. This would result in corneal pain non-treatable with aforementioned drugs. Anticonvulsants, opiates and topical local anesthetics can manage corneal neuropathic pain. The anticonvulsant Gabapentin (Neurontin) is an analog of gamma-aminobutyric acid (GABA), and its reliability in treating corneal pain is conflicting, mainly because of a lack of studies. Lichtinger et al. compared in a prospective randomized, double-blind, placebo-controlled study the efficacy of Gabapentin in the reduction of the corneal pain. They administrated gabapentin capsules (300 mg) in 20 patients and additional 20 patients received identical placebo capsules. They demonstrated that gabapentin reduced corneal pain during the first 24 h (P=.003), at postoperative day 1 (P=.002), between 24 and 48 h (P=.024), at postoperative day 2 (P=.018) and between 48 and 72 h (P=.001). Faktorovich et al. trying to prove the efficacy of topological opioid in the treatment of pain, concluded in a double-blind randomized prospective study that the administration of 0.5% morphine drops was an effective and safe method to control of post-PRK pain, and did not hamper epithelial healing or refractive outcomes. Topical local anesthetics include tetracaine, proparacaine, lidocaine and bupivacaine can be also used. Topical tetracaine has been documented to be successful in pain control management and does not produce delayed corneal healing times. However, it produces keratocyte toxicity and keratitis. Topical anesthetics should be used cautiously and for short-term treatments. Antidepressants are prescribed for neuropathic pain management elsewhere in the body. To date, no study has been published evaluating the effect of antidepressants for treating post-PRK corneal pain.

Conclusions

Photorefractive keratectomy disrupts corneal structure affecting epithelium, Bowman’s membrane, and anterior stroma. Corneal nerves are severed, which alters corneal integrity and function temporarily. The subsequent corneal wound healing is a complex process that is regulated by a variety of factors. A balance of peptides will determine the final outcome, and the presence of postoperative complications. Corneal wound healing process can be managed with several drugs to enhance regeneration and prevent corneal haze and pain after PRK. Further research in this field is required to completely understand post-PRK corneal regeneration in order to prevent complications, and provide outstanding visual outcomes.

Directions for Future Research

This review clearly states that corneal regeneration after PRK is not completely understood. The ongoing research in new drugs development, more efficient surgical techniques, and new imaging technologies are trying to answer some of the unresolved questions. Still, future research should be oriented to elucidate the following aspects:

- The long-term effects of keratocyte death and MMC application.
- Although corneal haze has been correlated to several factors, its origin is still unknown.
- The beneficial role of corticosteroid administration in corneal haze prevention.
- The causal factors of myopic regression.
- More studies using non-contact gas esthesiometer will help to better assess the time course of corneal sensitivity recovery.

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