



## The Effect of a New Rigid Gas-Permeable Contact Lens Design on Lactic Dehydrogenase Activity in Rabbit Tears

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**Key words:** contact lens periphery, lactic dehydrogenase, tears, diffractive relief, corneal metabolism, corneal hypoxia

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### Abstract

**Background:** Most corneal damage induced by contact lenses is due to interference with corneal oxygenation.

**Objective:** To investigate the effect on the rabbit cornea of a rigid gas-permeable contact lens with a newly designed periphery.

**Method:** We fitted New Zealand white rabbits (n=12) with RGP contact lenses that were identical in all respects except for the design of the periphery. In each animal, one contact lens had an innovative periphery consisting of a microscopic diffractive relief lathed on the back surface; the other contact lens was of a conventional design. The lenses were worn continuously for 7 days. During this experimental period and for 1 additional week we assessed the corneal damage by daily testing lactic dehydrogenase activity in the tears.

**Results:** On the last day of the experimental week and the first 3 days of the healing period, mean tear LDH activity was significantly lower in the eyes with the new contact lens design than in eyes with the conventional lenses.

**Conclusions:** The novel periphery design reduces corneal damage resulting from contact lens wear, as reflected by LDH levels in the tears. The new design probably facilitates the flow and exchange of tears under the contact lens, resulting in improved metabolism of the cornea. These findings may also prove applicable to soft contact lenses.

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Contact lenses are increasing in popularity as new materials and designs render them easier to fit and healthier and more comfortable to wear. Rigid gas-permeable contact

lenses are believed to interfere less than soft contact lenses with corneal metabolism. Nevertheless, long-term effects such as capillary infiltration of the peripheral cornea, corneal deformation, pinguecula formation, and giant papillary conjunctivitis are encountered in contact lens wearers after years of use [1]. These changes are due, at least in part, to the long periods of interference with normal corneal metabolism and the relative hypoxia of the cornea. Their severity may depend on one or more of several factors, such as the permeability of the material to oxygen and the fit of the lens to the cornea (steepness or flatness of the surface, centration over the cornea). An imperfect fit could impair adequate tear exchange behind the lens, thus affecting both oxygen supply and the removal of metabolic products. Although refractive changes (especially with-the-rule astigmatism) are usually related to the fit of the lens, they occasionally appear as a reaction of the cornea to lenses that prevent effective tear exchange [2].

In an attempt to minimize these long-term effects, considerable effort has been devoted to improving corneal compatibility and particularly to increasing oxygen permeability of the raw materials. Thus, the newer contact lenses are not only thinner, thanks to stronger and more stable materials, but they also have higher oxygen transmission capacities and their chemical components are more compatible with the eye. In spite of the fact that new contact lens designs are primarily aimed at improvement of comfort rather than corneal health, elliptical base curves and smoother blending of the different curves also refine contact lens fitting and the relationship of the lens to the cornea.

During human clinical trials with a new trifocal diffractive contact lens [3], some patients voluntarily reported that the lens felt very comfortable. On the assumption that the feeling of comfort is related to the diffractive design, we undertook to test the design in an animal model using tear lactic dehydrogenase as an indicator of corneal well-being.

RGP = rigid gas permeable  
LDH = lactic dehydrogenase

A variety of procedures, including measurement of aqueous humor acidification [4], corneal edema recovery [5] and corneal confocal microscopy [6], have been used to determine the effects of contact lenses on corneal health. Assessment of corneal damage in terms of LDH levels in tears has been described in a number of recent reports [7–10]. Tear LDH levels, measured in combination with morphological studies, have yielded stable and reproducible results [11]. The enzyme is well known in the diagnosis of cardiac ischemia and liver diseases where high serum levels are found. Tear LDH originates mostly in corneal epithelium [12] and its concentration is the highest of all the enzymes in tear fluid [13,14]. In the present study we used measurements of LDH activity in the tears to evaluate the effect of a new design of the RGP contact lens on rabbit corneal epithelium.

## Materials and Methods

Twelve New Zealand white rabbits weighing 3–4 kg were used. Examination of all the animals prior to contact lens fitting revealed no detectable ocular disease. The rabbits were handled according to the ARVO Resolution on the Humane Use of Animals in Vision Research.

Both eyes of each rabbit were fitted with lenses that were identical in terms of material (Boston ES), base curve (7.2–7.8 mm), diameter (12.5 mm), central thickness and optical power (plano). All lenses had an elliptical base curve geometry with identical eccentricity. The only difference between the lenses in the two eyes was in the design of the periphery: the rigid monofocal lens had a conventional periphery, while the rigid monofocal diffractive lens had a periphery designed to improve tear flow under the lens. We used a novel microscopic diffractive relief, which had originally been designed and lathed on the back surface of RGP contact lenses in order to manufacture a trifocal contact lens [3]. In the present study we were interested in the hydrodynamic features of the lens, not its optical performance. Accordingly, we lathed the microscopic relief concentrically on the peripheral back surface of the contact lens without impinging on the optic zone, thus avoiding interference with the optical features of the lens.

Initial fitting was done with a trial set, care being taken to fit both lenses in each rabbit so as to obtain the same fluorescein picture, indicating alignment fit. In the four cases in which this was not accomplished both lenses were steep (one case) or both were flat (three cases). If lenses with the same base curve did not show the same fluorescein pattern the rabbit was excluded from the study.

After the initial fitting, lenses remained in the eyes for 7 days. At the end of this period they were gently removed with a vacuum contact lens remover, and daily clinical ocular examinations of the animals were continued for a further week. To exclude the possibility of diurnal variation [7], tear samples (4–5  $\mu$ l) were collected between 2:00 and 3:00 p.m., before the ocular examination,

from both eyes of each rabbit. Tear samples were collected in 10  $\mu$ l glass microcapillary tubes (Blaubrand, Intramark, Germany) from the inferior meniscus prior to lens insertion and every 24 hours during contact lens wear. Tear collection was performed very gently, with not even minimal disturbance to the animal's eyes, therefore no anesthetic was required during the procedure. The contact lenses were not removed during tear collection and care was taken to avoid touching the cornea, contact lens or conjunctiva, so as to prevent cellular damage that might artifactually increase LDH levels. On the 7th day of lens wear, tear samples were collected and the lenses removed. Further samples were then collected on 1, 2, 3 and 7 days after removal of the lenses. Tears (2  $\mu$ l) were diluted with 98  $\mu$ l of 50 mM phosphate buffer (pH 7.5) and the diluted samples were subjected to LDH assay.

LDH was measured spectrophotometrically using a Monarch 2000 analyzer (USA). The assay is based on conversion of the co-enzyme nicotinamide adenine dinucleotide (NAD) to its reduced form (NADH) at 340 nm, upon the conversion of lactate to pyruvate [15]. Mean tear LDH activity was calculated separately each day for both types of lenses. A control group of two rabbits (four eyes) without contact lenses had their LDH levels similarly determined over a 2 week period. The LDH measurements were performed in a masked fashion.

Statistical analyses of the data were performed using the ANOVA test. Results were considered significant when  $P < 0.05$ .

## Results

### Clinical evaluation

Daily slit-lamp biomicroscopic examination with and without fluorescein showed increasing corneal damage when the contact lenses were worn, although the degree of severity varied among animals. Clinical examinations were performed without touching the contact lenses in order to minimize the risk of additional corneal damage during contact lens removal and/or insertion. We observed progressive central corneal erosions, blood vessel infiltration of the peripheral cornea, conjunctival and especially limbal hyperemia, and mucoid discharge. These findings are expected with contact lenses that, although not designed for extended wear, are worn for prolonged periods.

### Measurement of LDH activity

Prior to fitting the contact lenses, mean LDH activity in tear fluid was  $1,525 \pm 1,375$  U/L in eyes that subsequently wore the newly designed lenses (RMD) and  $2,104 \pm 2,180$  U/L in eyes that wore RM contact lenses. The large standard deviations are probably due to the variability inherent in the tear sampling method.

The difference between the two experimental groups was not significant ( $P = 0.16$ ), indicating that any differ

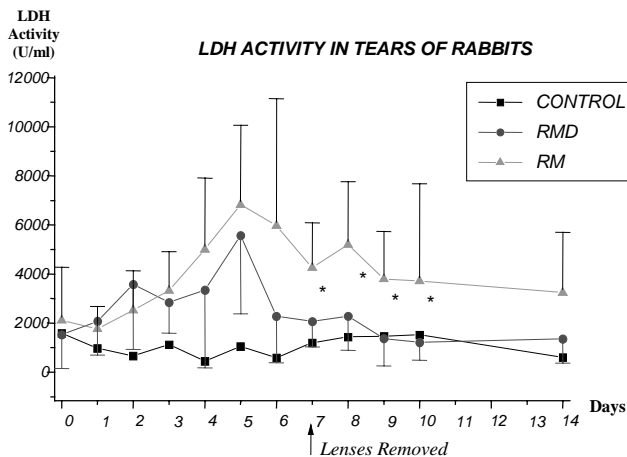
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RMD = rigid monofocal diffractive  
RM = rigid monofocal

**Table 1.** Mean lactic dehydrogenase activity in tears of rabbits by lens design

	Rigid monofocal diffractive lenses	Rigid monofocal lenses	Control (no lens)
Before study	1,525 ± 1,375 U/L	2,104 ± 2,180 U/L	
During study	2,546 ± 1,211 U/L	4,148 ± 1,429 U/L	905 ± 403 U/L*

\* Values in the control group were obtained without contact lenses, before and during the study.



**Figure 1.** LDH levels in tears of rabbit eyes using newly designed and conventional contact lenses for 7 days and in controls. RMD = mean LDH levels in eyes using rigid monofocal diffractive lenses, RM = mean LDH levels in eyes using conventional rigid monofocal lenses. \* Statistically significant (ANOVA)

**Table 2.** Mean LDH levels in tears of rabbits wearing newly designed (RMD) and conventional (RM) contact lenses for 7 days

Day of study**	Rigid monofocal diffractive lenses		Rigid monofocal lenses		P value*
	Mean	SD	Mean	SD	
0	1525	1375	2104	2180	.464
1	2081	1380	1763	920	.537
2	3584	2652	2525	1613	.378
3	2847	1258	3328	1589	.517
4	3350	3169	5000	2912	.299
5	5571	3193	6825	3246	.512
6	2278	1888	5972	5177	.097
7	<b>2070</b>	<b>1038</b>	<b>4261</b>	<b>1833</b>	<b>.014</b>
8	<b>2283</b>	<b>1391</b>	<b>5200</b>	<b>2572</b>	<b>.007</b>
9	<b>1370</b>	<b>1119</b>	<b>3802</b>	<b>1931</b>	<b>.002</b>
10	<b>1215</b>	<b>726</b>	<b>3719</b>	<b>3968</b>	<b>.049</b>
14	1354	983	3239	2468	.107

\* Statistically significant results are printed in bold letters.

\*\* Days 8, 9, 10 and 14 represent days 1, 2, 3 and 7, respectively, after contact lens removal.

ences found between them during or after contact lens wear was attributable to the lenses themselves. Mean tear LDH activity during the 2 week period of the study (baseline excluded) was significantly lower in eyes wearing RMD lenses than in eyes wearing RM lenses ( $P=0.014$ ). As expected, significantly lower mean LDH activity values were obtained in the control (no lens) group ( $P<0.001$ ). The results are shown in Table 1.

Mean tear LDH activity in the three groups of eyes over the 2 week study period is shown in Figure 1 and Table 2. It can be seen that from the third day, tear LDH levels were lower in eyes wearing RMD lenses than in eyes wearing the regular lenses. On the 7th day the difference became statistically significant [Table 2].

We continued to collect tear samples during the first 3 days following lens removal and also on the 7th day (the 14th day of the study). Tear LDH activity during the first 3 days of the recovery period was significantly lower in the RMD contact lens group than in the group wearing RM lenses [Table 2]. In the former group LDH levels returned to baseline after 2 days. In contrast, 1 week after lens removal LDH activity was still higher than baseline in the RMD lens group, although the difference between the two groups at that stage was not statistically significant ( $P=0.16$ ).

## Discussion

In this study we examined the effect on rabbit corneal health of RGP contact lenses with a newly designed periphery. The novel design incorporates a diffractive relief, that is lathed on the peripheral back surface of the contact lens and is assumed to improve tear flow dynamics under the contact lens. We suggest that either of two mechanisms or a combination of both may explain the results. One possibility is that the microrelief reduces resistance to tear flow under the contact lens; the other is that the capillary flow of tears under the contact lens is enhanced. Whatever the explanation, increased tear flow under the lens can be expected to decrease corneal hypoxia and improve corneal metabolism by permitting metabolites to pass more freely towards and away from the center of the cornea. Accordingly, chronic changes resulting from prolonged contact lens use might be diminished.

The measurement of LDH activity levels in tears, used here to evaluate the physiological effect of contact lenses, has been employed in a number of studies to determine what effect contact lenses made from materials with different oxygen transmissibility characteristics (Dk values) had on the cornea. These studies indicate that the higher the LDH level, the more serious the corneal damage [7–11]. The large variation in enzyme levels that we observed at baseline and during the experimental period was also encountered in the previous studies. Whereas other studies using rabbits to evaluate contact lens designs or materials removed the nictitating membrane, we chose not to. Our reasons were based on the

observation that contact lenses were well tolerated and were not dislodged even when the membrane was left intact, and on the assumption that any interference with the natural anatomy of the rabbit eye might induce changes that would later be difficult to interpret.

Although this study was performed with rabbits, we believe that the reaction in human corneas is likely to be similar since a study of human subjects demonstrated a strong correlation between tear LDH activity and pathological corneal effects of contact lens use [8].

In the present study, serious corneal insults were produced in rabbits by both types of RGP contact lenses worn continuously for 7 days. Although the changes occurring in eyes after long-term contact lens wear were different from the short-term effects in our study, we believe that our findings may be applicable for prolonged wear. As shown in Figure 1, there was a striking difference in LDH activity between the two groups in favor of the newly designed contact lenses: namely, lower levels in the RMD group from the third day of the study until the termination of the experiment on the 14th day. The results became statistically significant ( $P=0.014$ ) on day 7 (the last day of contact lens wear), although by day 6 they were already close to significance ( $P=0.097$ ). This might be explained by the fact that tear fluid containing high levels of LDH were 'trapped' under the contact lens and it was only on day 7 that the leak of enzyme showed significantly higher values in the regular lens group. It appears from Figure 1 that enzyme levels declined after reaching a peak on the 5th day of contact lens wear. Theoretically, tear enzyme levels in such experiments should increase with time, but we consistently found with both types of contact lenses that enzyme levels were higher on the 5th than on the 6th and 7th days. This finding further supports our opinion that tear fluid was trapped under the contact lenses and could not be sampled, resulting in erroneously low LDH readings. We chose not to remove the contact lenses during tear collection, as the manipulation might have caused further corneal damage and thus produce misleading results.

LDH levels after lens removal demonstrated that rabbit eyes wearing the newly designed contact lenses for 7 days recovered after 2 days, with their tear LDH levels returning to baseline. In eyes that had worn the regular contact lenses, the enzyme levels did not return to baseline even after 3 days and decreased very little by the 7th day. Both types of contact lenses were manufactured from the same raw material and their parameters were identical, including central thickness and vertex power. It seems reasonable to conclude that the corneal recovery after removal of the newly designed contact lenses was quicker because the corneal damage was less severe than with conventional contact lenses. We believe that this lower severity is attributable, at least in part, to decreased corneal damage and improved corneal metabolism in eyes wearing the

RMD lenses. It is thus possible that the new design permits enhancement of tear flow between the cornea and lens. Although the precise mechanism has yet to be elucidated, the results of this experiment suggest that the capacity of the cornea to cope with long-term contact lens use is improved when a microscopic diffractive relief is lathed on the contact lens periphery.

RGP contact lenses are generally considered not to harm the ocular surface. It is obviously desirable that the impact of contact lenses on eye tissue should be as mild as possible. The newly designed lenses described in this study appear to represent a step in this direction. The design may be relevant also for soft contact lenses and we are in the process of examining this possibility. If it proves beneficial, this will be an even more important finding in view of the deleterious effects of soft lenses on the cornea.

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