

Figure 2.--The IC spaces in a renal papilla, with an opened pelvis, that was microinjected with ~50 nl of a 10% ferritin solution for about 1 min prior to the snaring of the papilla. A and B show the same cell at two different magnifications. These cross sections were taken 250 μ m from the tip of the papilla. The small black ferritin granules are seen to be concentrated in the IC spaces. C and D also show one cell at low and at high magnification, respectively. These cross sections are 300 μ m from the tip of the papilla. Again the IC spaces are filled with ferritin granules.

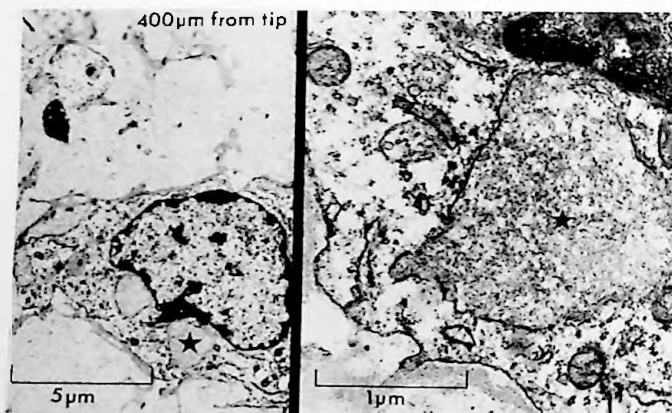


Figure 3.--The IC spaces in a renal papilla injected through the intact renal pelvic wall with approximately 50 nl of a 10% ferritin solution. The injection lasted 30 sec, the papilla was snared and fixed 15 sec after the end of the injection. The figure to the right is a higher magnification of the IC space shown in the figure to the left. The ferritin granules were sparse in this section as well as in the sections below. This may indicate that the ferritin has been rather effectively removed in this papilla. The removal has probably been facilitated by the peristaltic contractions of the intact renal pelvic wall.

The IC of the inner medulla have not previously been implicated in fluid transport. They are known to be the site of prostaglandin synthesis. Thus, in both rats and rabbits about half of the medullary synthesis of PGE_2 takes place in the IC. Churchill et al (Fed. Proc. 41: pX, 1982) reported that in antidiuretic rats the IC have almost 70% higher electrolyte concentrations than the surrounding interstitium and collecting duct cells. At this time it is not known if this high electrolyte concentration is involved in moving fluid into the IC spaces. An important question to be answered is whether the IC spaces connect with the lymphatics at the border between the outer medulla and cortex. Our current experiments are addressing this point.

CORNEAL WOUND HEALING IN SCULPIN AND SHARK

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Corneal epithelial wound healing in mammalian species has been quantified and occurs by epithelial cell

sliding and mitosis. This problem has great clinical importance since studies of corneal wound healing will aid in the elucidation of the pathophysiology of corneal epithelial erosions and persistent epithelial defects. Past studies have shown that the mammalian cornea is capable of re-epithelializing at a rate of 0.7 to $1.2 \text{ mm}^2/\text{hr}$ and this rate can be increased with growth factors and retinoic acid. The purpose of this study was to compare the rate of corneal wound healing in the sculpin, which has a swelling cornea, and the shark, which has a non-swelling cornea. These corneas differ structurally in their swelling properties and in their adaptation to their osmotic environment (Edelhauser et al., Fed. Proc. 39:3213, 1980).

Sculpin (*Myoxocephalus octodecimspinosus*) and dogfish shark (*Squalus acanthius*) were anesthetized with MS222. A central corneal epithelial wound was made on sculpin and shark eyes by scraping the epithelium within a 6.5 mm trephine mark. These wounds were 30% of corneal area in the sculpin and 25% in the shark. A 60% wound was also made in one group of sculpins by scraping within a 1.1 cm trephine mark. The corneal wounds were stained with sodium fluorescein ophthalmic strips moistened with sea water, and the excess fluorescein was washed from the cornea with sea water. The corneal wounds were viewed with long-wave UV and photographed with Kodak Tri-X black and white film using a Wratten 47B filter on the electronic flash and a Wratten 12 filter on the camera lens. Slides were projected onto paper and the wound perimeters were traced. The areas of the tracings were determined by hand planimetry. After the initial photographs, the wounds were stained and photographed at 3, 6, 9, 12, 24, 28, 32 and 48 hours.

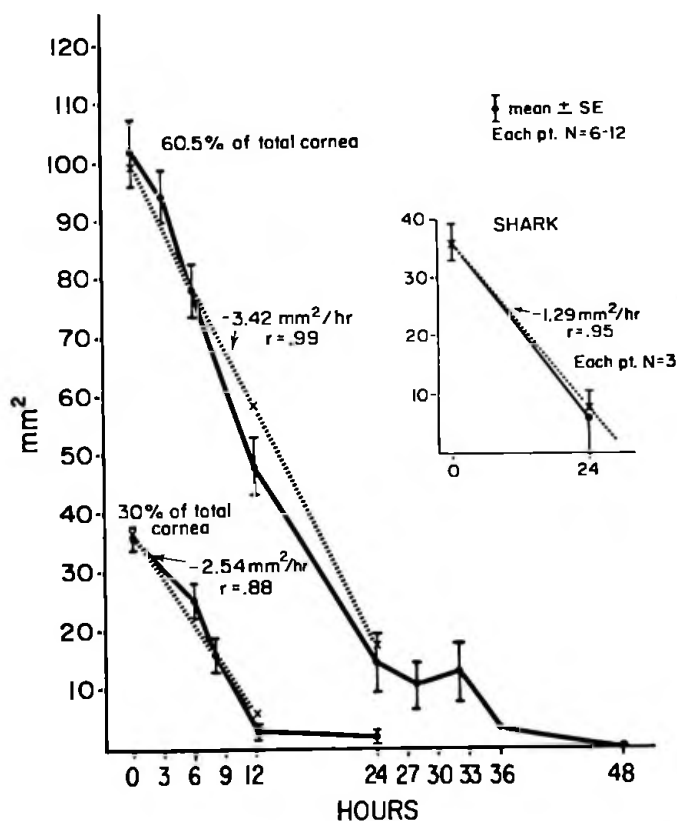


Figure 1.--Corneal wound healing following epithelial scraping in sculpin and shark. The rate of healing and correlation coefficient (r) are listed next to the calculated regression line.

The initial wounds varied from 29 to 40 mm^2 for those wounds scraped within the 6.5 mm trephine and between 130 to 193 mm^2 within the 1.1 cm trephine mark. During scraping the knife blade occasionally crossed the trephine

mark resulting in wound size variation. There was, however, no correlation between initial wound size and healing rate within each group. This observation has been reported by Ho et al (Invest. Ophthalmol. 13:804, 1974).

The average healing rates of the sculpin cornea were $2.54 \text{ mm}^2/\text{hr}$ and $3.42 \text{ mm}^2/\text{hr}$, respectively, for 30% and 60% corneal wounds. In shark the corneal wounds healed at $1.29 \text{ mm}^2/\text{hr}$ (Figure 1). By comparison, the results from this study show that the sculpin corneal healing rate is two- to threefold higher than those reported for many mammalian species (Ubels et al. J. Toxicol.--Cut. & Ocular Toxicol. 1:125, 1982). Following topical treatment of standard corneal wounds in rabbits with epidermal growth factor (EGF), the maximum corneal healing rate has been reported to be $1.29 \text{ mm}^2/\text{hr}$ (Ho et al., Invest. Ophthalmol. 13:804, 1974), and recently it has been reported that an Eye Derived Growth Factor (EDGF) will increase the corneal healing rate to $1.45 \text{ mm}^2/\text{hr}$ (Thompson et al., Exp. Eye Res. 34:191, 1982). The value we measured in shark cornea is similar to these stimulated values. It is interesting to note that, in order for these marine animals to survive in their environment, it is essential that vision be preserved, and this study illustrates the high rate of corneal re-epithelialization that these species possess in order to maintain their vision and prevent stromal edema following a corneal laceration. This research was supported in part by NEI Grant EY-00933. Dr. Edelhauser is a 1982 Research to Prevent Blindness, Inc., Olga K. Weiss, Research Scholar.

THE EFFECT OF HYPERBARIC OXYGEN ON THE ELECTRORETINOGRAM OF MARINE TELEOSTS AND ELASMOBRANCHS

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INTRODUCTION--The mammalian retina is highly susceptible to oxygen toxicity; however, it has been reported that the electroretinogram (ERG) recorded in vitro from the retinas of rainbow trout (Salmo gairdneri) and goldfish (Carassius auratus) is unaffected by exposure of the retina to O_2 at 3800 mmHg for up to 6 hours (Ubels et al., Comp. Biochem. Physiol. 57A:29, 1977). This resistance to oxygen toxicity is probably an adaptation to the high P_{O_2} ($> 200 \text{ mmHg}$) generated by the counter-current O_2 multiplier of the choroidal rete mirabile found in most teleost fishes. Since this phenomenon has only been studied in fishes with well-developed retes and elevated retinal P_{O_2} , the present study was designed to test this hypothesis by recording the in vitro ERG from the retinas of several marine fishes with varying degrees of rete development and retinal P_{O_2} , during exposure to hyperbaric oxygen.

MATERIALS AND METHODS--Three teleost species were used: the flounder (Pseudopleuronectes americanus) which has a large rete and elevated retinal P_{O_2} ($> 200 \text{ mmHg}$), the goosefish (Lophius americanus) which has a small poorly developed rete and low retinal P_{O_2} ($< 90 \text{ mmHg}$), and the eel (Anguilla rostrata) which has no rete mirabile. ERG's were also recorded from the eyes of two elasmobranchs, the spiny dogfish (Squalus acanthias) and the skate (Raja erinacea) which have no rete and a retinal P_{O_2} reported to be less than 20 mmHg (Wittenberg and Wittenberg, Nature 194:106, 1962). Eyecups were prepared by removing the cornea, iris and lens from enucleated eyes. The ERG was recorded simultaneously from paired retinas under 100% O_2 at 3800 mmHg and at 760 mmHg (control) as previously described (Ubels et al., Comp. Biochem. Physiol. 57A:29, 1977). The a-wave and b-wave amplitudes were measured directly from the oscilloscope screen, and the ERG's were photographed. All experiments were conducted at 15° to 17°C .

RESULTS AND DISCUSSION--As predicted, based on results of previous studies of trout and goldfish, the flounder ERG was not attenuated by 6-hour exposure to HBO and, in fact, was somewhat enhanced (Table 1; Figure 1A). The goosefish retina, with its poorly developed rete and lower P_{O_2} is susceptible to oxygen toxicity as demonstrated by a 50% reduction in b-wave amplitude after a 6-hour exposure to HBO (Table 1; Figure 1B). The eel does not fit the hypothetical pattern since HBO had no effect on the ERG in spite of the lack of a normally elevated retinal P_{O_2} in this animal. This observation agrees with previous data on the effect of HBO on retinal Na^+/K^+ ATPase in the eel (Ubels et al., Bull. MDIBL 21:53, 1981).