MEASUREMENT OF STROMAL SWELLING PRESSURE IN ELASMOBRANCH AND SCULPIN CORNEA

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The elasmobranch cornea is a non-swelling tissue with the swelling physically limited in these species by the sutural fibers which interconnect adjacent stromal lamellae (Goldman et al., Invest. Ophthalmol. 6:574, 1967). The measured swelling pressure of the cornea is zero at normal hydration (Talpin et al., Exp. Eye Res. 8:429, 1969), and no active transcorneal ion transport is found in this tissue (Fischer et al., Exp. Eye Res. 25:149, 1977). Hence, this tissue need not expend metabolic energy for active deturgescense. Metabolic studies have shown that this cornea exhibits a minimal rate of glucose oxidation (Geroski et al., Exp. Eye Res. 32:133, 1981). The cornea of the longhorn sculpin, a marine teleost, is more closely related to the corneas of higher vertebrates. It is a swelling tissue showing active transcorneal ion transport (Fischer et al., Exp. Eye Res. 25:149, 1977) and significantly higher metabolic activity than the elasmobranch cornea (Geroski et al., Exp. Eye Res. 32:133, 1981). The cornea of the sculpin is divided into an anterior and posterior strama, and past studies (Edelhauser et al., Bulletin 21:26, 1981) have shown that both stromas have similar swelling properties in Ringers and deionized water.

The objective of this study was to compare the swelling pressure and the loss of stromal proteoglycans during swelling of the shark stroma and the anterior and posterior stroma of the sculpin to the measured swelling pressure of the rabbit cornea, a representative mammalian species with a known swelling pressure of 60 mmHg.

Corneas were excised from enucleated eyes of dogfish shark, sculpin, and rabbits. The epithelium and endothelium were removed with a Gill knife, and a 6.5-mm stromal button was trephined. The button was immediately weighed and dehydrated at room temperature in a dessicator to a hydration value below the initial weight. The stromal buttons were then placed between two 7.0-mm diameter sintered stainless steel discs, and the swelling pressure was measured according to the method of Hedbys and Dohlman (Exp. Eye Res. 2:122, 1963). A volume of 400 µl of deionized H_2O or Ringers was added to the stromal well, and the swelling pressure was recorded over a 30-minute period (Figure 1). After the swelling pressure was determined, the stromal buttons were

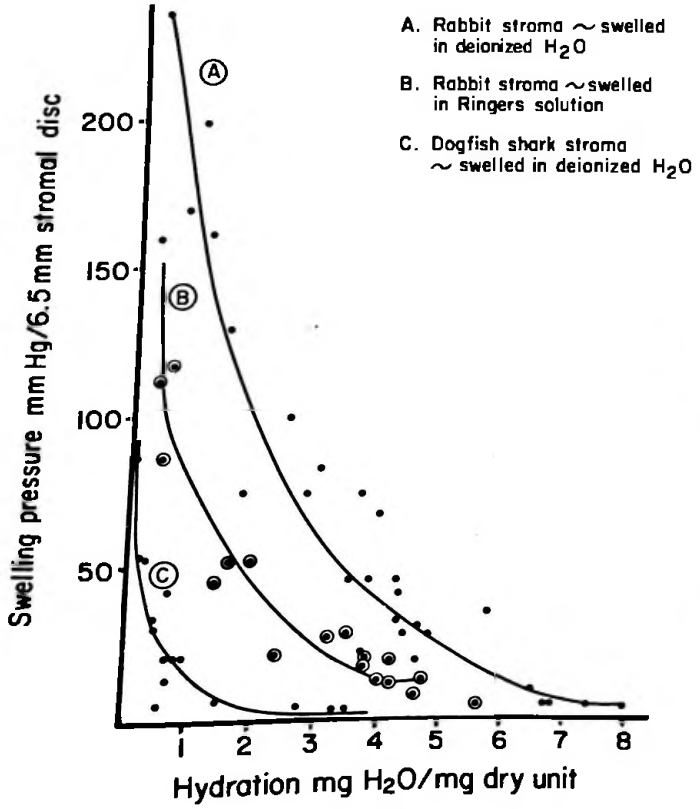

Figure 1.--Swelling pressure as a function of hydration for rabbit and shark corneal stroma.

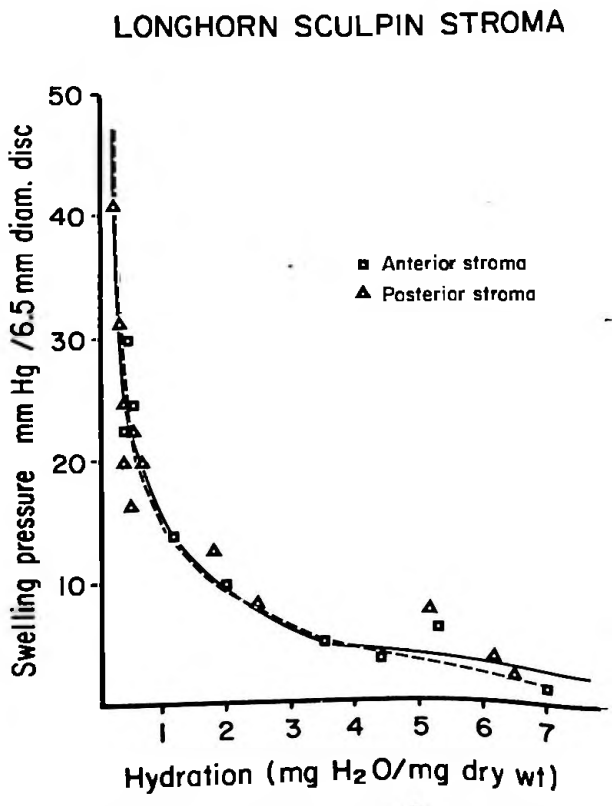

Figure 2.--Swelling pressure as a function of hydration for the anterior and posterior corneal stroma of the sculpin.

removed, blotted dry, weighed, dried at 120°C for 24 hrs, and reweighed. The stromal hydration was then determined from the wet and dry weight (Table 1). Stromal buttons were also incubated at the various hydration values

Table 1.--Normal (mean ± SE) Stromal Hydration and the Dry Weights of 6.5-mm Diameter Stromal Buttons from the Cornea of Rabbit, Shark, and Longhorn Sculpin

Animal	Normal Stromal Hydration (mg H_2O/mg dry wt)	Dry wt (mg) 6.55-mm diameter button
Rabbit	3.5 ± 0.1 (3)*	3.5 ± 0.1 (10)
Shark (Adult)	3.4 ± 0.1 (3)	1.1 ± 0.1 (10)
Sculpin		
Anterior	6.4 ± 0.2 (4)	1.3 ± 0.2 (10)
Posterior	5.0 ± 0.2 (3)	0.8 ± 0.2 (10)

*( ) number of samples.

in 175 µl of deionized water for 30 minutes. At the end of the 30-minute incubation period, the solution was assayed for total proteoglycan content (Gold. Analyt, Biochem. 99:183, 1979).

The swelling pressure of the rabbit stroma at normal hydration (3.5 mgH_2O/mg dry wt) is 60 mmHg in deionized water and 25 mmHg in mammalian Ringers. When the stromal hydration is decreased, the stromal swelling pressure increases (Figure 1). By comparison, the shark stroma has a swelling pressure of less than 10 mmHg at normal hydration (3.4 mg H_2O/mg dry wt) which can increase to 60 mmHg when the stromal hydration is decreased. These values are similar to those reported by Tolpin et al (Exp. Eye Res. 8:429, 1969). Associated with this swelling pressure was a loss of stromal proteoglycans (22.7 µg/6.5-mm button/15 min) to the deionized water by the rabbit stroma. This value was greatly reduced in the shark cornea (7 µg/6.5-mm button/30 min). Cremer-Bartels et al. (Jap. J. Ophthalmol. 27:305, 1983) have also shown that the loss of glycosaminoglycans and glycoproteins is greatly reduced in the shark cornea in comparison to the rabbit tissue. These observations could be correlated to the sutural fibers and the collagen make-up of the shark cornea.

The swelling pressure of the anterior and posterior stroma of the sculpin is also less than the rabbit but comparable to the shark at normal hydration (6.4 and 5.0 mg H_2O/mg dry wt, respectively), which can increase to 30-40 mmHg when the hydration value decreases (Figure 2). Despite this low swelling pressure there was a considerable loss of the stromal proteoglycan by the anterior stroma (22.7 µg/6.5-mm button/30 min) and a lesser amount by the posterior stroma (8.7 µg/6.5-mm button/30 min).

These results suggest that the corneal stromal swelling characteristics of the shark, sculpin, and rabbit are varied and controlled by the structural make-up of the stroma. The comparison between sculpin and rabbit stromal swelling and proteoglycan loss may partially account for the reported nonuniform swelling observed in edematous mammalian corneas (Kikkawa and Hirayama, Invest. Ophthalmol. 9:735, 1970; Lee and Wilson, Current Eye Res. 1:457, 1981). This study was supported in part by National Eye Institute Grant EYOO933 and by NIEHS Aquatic Biomedical Center Grant ESO1985.

TAURINE TRANSPORT BY THE FLOUNDER INTESTINE

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Taurine is found in relatively high concentrations (> 20 mM) in the tissues (heart, kidney, rbc, liver) of a variety of marine fish and is important in the regulation of cell volume. There is no evidence of metabolic pathways for the synthesis of taurine in any of the fish tissues examined (King et al., J. Exp. Zool. 212:78-86, 1980).