DEVELOPMENT OF THE CORNEA IN THE EYE OF THE CLEARNOSE SKATE (RAJA EGLANTERIA)

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Most descriptive and experimental data about the steps involved in corneal development have come from studies of the embryonic chick. In that species, formation of the definitive stroma is preceded by formation of a distinct orthogonal gridwork of collagen fibrils (primary stroma) by the corneal epithelium; the mature form of the predominant corneal stroma proteoglycan, keratan sulfate proteoglycan (KSPG), appears after ~1/3 of embryonic development has occurred. The initially opaque-translucent cornea only attains full transparency just before hatching. Development of the mammalian cornea differs from this pattern in a number of respects, depending on the type of mammal.

We have begun to characterize the steps in corneal development in a vertebrate that offers several advantages over embryos of birds and mammals. Embryos of the clearnose skate, Raja eglanteria, require only room temp. (20-22°C) slowly circulating sea water for normal development to proceed to hatching after ~82 days. From Day 27 onward, the embryos are more accessible experimentally than embryos of birds or mammals. These embryos are large and robust, resembling chick embryos in overall morphology for much of early development.

In work described here, three types of observations were made: first, corneal transparency was assessed by ability to visualize internal eye structures through the cornea itself. Second, embryos were fixed at various stages, embedded, sectioned, and stained with a monoclonal antibody that reacts with the sulfated KS epitope diagnostic of mature KSPG molecules in the corneas of chicks and humans. Third, fixed embryos were sectioned and examined by transmission electron microscopy. Results: as assessed to this date, the data indicate that: 1) the cornea is crystal-clear transparent from its earliest stages; 2) KSPG is absent from the cornea even at stages half-way through embryonic development, although this proteoglycan is present definitively in some cartilaginous structures of the same embryos, as well as in the adult cornea of a related species (R. erinacea); 3) Collagen fibrils are present in orthogonal gridwork in the stroma even early in development. Whether this stromal matrix is a primary stroma analogous to that of the bird remains to be determined.

Conclusions: A mature, highly-sulfated form of KSPG may not be necessary in the stroma as a prerequisite 1) for corneal transparency (such a molecule normally is present when the embryonic chick cornea begins to become transparent), or 2) for formation of very early, orthogonally ordered plies of collagen fibrils (such a molecule normally is absent during the formation of the primary stroma of the embryonic chick cornea).

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