

by Olsen and Kent (Cell Tiss. Res. 209: 49-63, 1980). Our findings of a beta-adrenergic control of both branchial resistance and pattern of flow is to be contrasted with the general pattern in teleosts where afferent pressure is controlled by a beta-adrenergic site, presumed to be on the prelamellar arteriole, while perfusate flow into the dorsal aorta is controlled by alpha-adrenergic sites, presumably at the postlamellar arteriovenous anastomoses (Payan and Girard, Am. J. Physiol., 232:1718-1723, 1977; Claiborne and Evans, 1980, op. cit.).

When the acetylcholine agonist carbachol ( $5 \times 10^{-6}$  M) was added to the perfusate the afferent pressure increased by  $7.5 \pm 4.5$  torr (3) some 12 to 20 minutes after the drug reached the tissue. The response was reversible. Perfusion with atropine ( $5 \times 10^{-5}$  M) alone did not alter either parameter. Thus it appears that at least gill resistance is also controlled by muscarinic, cholinergic inputs.

Our data demonstrate that the isolated, perfused head of the dogfish "pup" is a viable preparation for the study of the hemodynamics of the branchial vasculature. It has shown clearly that gill resistance can be increased by stimulation of alpha-adrenergic or muscarinic-cholinergic receptors or decreased by stimulation of beta-adrenergic receptors. In addition, the pattern of blood flow is controlled by beta-adrenergic sites. Usefulness of this preparation for studies on branchial transport mechanisms of elasmobranchs await further experiments, but these preliminary hemodynamic investigations indicate that this may be a suitable vehicle for such studies. This research was supported by NSF PCM 81-04046.

#### THE TRANSEPIHELIAL POTENTIAL ACROSS THE GILLS OF THE ISOLATED, PERFUSED HEAD OF MYOXOCEPHALUS OCTODECIMSPINOSUS

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Recently, we have shown that the isolated, perfused head preparation (IPHP) of the long-horned sculpin, M. octodecimspinosus, exhibits a long term viability and response to epinephrine (Claiborne and Evans, Bull. MDIBL 19:96-101, 1979; J. Comp. Physiol., 138:79-85, 1980), and a unidirectional NaCl efflux which may be via separate rate-limiting pathways (Claiborne and Evans, Bull. MDIBL 20:50-52, 1980; Marine Biol. Letters 2:123-130, 1981). Since the assessment of transepithelial potential (TEP) changes are important during branchial ion transport experiments (see review by Evans, Amer. J. Physiol. 7(2):R224-R230, 1980), the present study delineates the effects of external ion substitutions on the TEP across the gills of the sculpin IPHP.

The IPHP of M. octodecimspinosus was prepared as described previously (Claiborne and Evans, 1980, J.C.P., op. cit.). Transepithelial potential (TEP) measurements were accomplished by inserting 4% agar Ringer's bridges into the afferent perfusion line and the peritoneal cavity of the IPHP. The bridges were also connected to a voltmeter (Keithley, Model 616) via two matched calomel electrodes. For comparison purposes, the in vivo TEP in the sculpin was measured according to the methods of Evans, et al. (J. Exp. Biol. 61:277-283, 1974).

Table 1.--Comparison of TEPs in M. octodecimspinosus during seawater ion substitutions in vivo and in vitro

	seawater	-Na	seawater	-Cl
in vivo (N = 6)	$7.15 \pm 0.64$	$-0.78 \pm 1.43$ ( $P < 0.001$ )	$5.47 \pm 0.91$	$8.68 \pm 0.84$ ( $P < 0.01$ )
IPHP (N = 4)	$9.28 \pm 2.29^*$	$1.03 \pm 1.49$ ( $P < 0.01$ )	$8.95 \pm 2.38^*$	$8.34 \pm 1.98$ (NS.)

All values represent TEP in mV; Mean  $\pm$  S.E. with the serosal side of the gills taken as reference. Comparisons of differences between seawater and -Na or -Cl seawater were made utilizing paired differences and Student's t-test.

\*A comparison of average seawater TEPs in vivo and in vitro indicated no significant difference between these values.

The TEP of the IPHP in seawater was  $7.7 \pm 1.4$  mV ( $N = 7$ ), while that recorded in vivo was  $7.2 \pm 1.6$  ( $N = 6$ ). The similarity between these values may suggest that the IPHP exhibits the same overall differential ion permeability and electrogenic ion transport as in vivo. Table 1 shows the effect of Na<sup>-</sup> and Cl<sup>-</sup>-free seawater on the TEP in vitro and in vivo. Na-free seawater depolarized the TEP of the sculpin to a much greater extent than the hyperpolarization by Cl-free media, in both the IPHP and the whole animal. This is an indication that the gills of the IPHP of the sculpin (and in vivo) possess a higher permeability to Na than to Cl. This is not surprising since the branchial epithelium of several other teleosts which are serosally electropositive exhibit a Na/Cl permeability ratio greater than 1 (see review by Kirschner, *Am. J. Physiol.*, 7(2):R219-R223, 1980). Cl-free seawater did not effect the TEP of the IPHP, but hyperpolarized the TEP of the whole animal. This suggests that the IPHP retains a lower Cl permeability than in vivo, thereby limiting the rate of Cl efflux from the preparation. We have shown previously that this is indeed the case, in that the rate of isotopic Cl efflux from the IPHP appears to be below in vivo rates (Claiborne and Evans, 1981, *op.cit.*), though the underlying reasons for the Cl permeability changes in the IPHP of the sculpin are yet to be resolved.

To date, the TEP across an isolated head preparation has not been reported in the literature. This parameter is a good test of the viability and branchial permeability of the IPHP and should provide useful information when monitored concurrent with gill ion transport studies. This research was supported by NSF grant PCM 81-04046 to DHE.

#### 12 INHIBITION OF CHLORIDE SECRETION BY BaCl<sub>2</sub> IN THE RECTAL GLAND OF THE SPINY DOGFISH, SQUALUS ACANTHIAS

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Soluble barium salts are known to produce poisoning, fatal if untreated, that is characterized by hypokalemia and muscle paralysis. The hypokalemia is thought to be due to the capacity of barium to decrease the efflux of potassium out of cells, although inhibition of both influx and efflux of potassium has been noted. The capacity of barium to reduce passive efflux of potassium has been used experimentally to probe transport characteristics of different epithelia. Inhibition of potassium efflux should result in depolarization of the cell membrane with consequent effects on the membrane transport of other ions. We have postulated a model for the secretion of chloride by the rectal gland in which the negative intracellular potential facilitates the movement of chloride across the luminal cell membrane into the duct. Barium salts, by decreasing the membrane potential should inhibit the secretion of chloride by the rectal gland. In the experiments reported here barium chloride was added to the perfusate of isolated rectal glands to test this hypothesis.

Rectal glands were perfused as previously described (Silva et al., *Am. J. Physiol.* 233:F298, 1977), except that the perfusate was prepared without sulfate, to avoid precipitation of barium sulfate. Barium chloride was added to the perfusate at concentrations ranging from  $10^{-4}$  M to  $5 \times 10^{-3}$  M. The results are summarized in Table 1. Barium inhibits chloride transport by the rectal gland in a dose dependent manner. No effect is seen at  $10^{-4}$  M, the lowest concentration used, and inhibition was most marked (82%) at a concentration of  $5 \times 10^{-3}$  M. Since the latter was the highest concentration used it is not known whether that is the maximal inhibition achievable. The effects of barium on chloride secretion are completely reversible as shown in the last row of the Table where, upon return to a perfusate that does not contain barium, chloride secretion returned to normal. Electrical potential difference measured across the gland is also inhibited by barium in a dose dependent way. The reduction in the electrical potential across the gland correlates well with the reduction in chloride secretory rate as evidenced by a correlation coefficient of  $.84$   $p < .05$ . As was the case with chloride secretion, the electrical potential difference across the gland returned to normal when the barium was removed from the perfusate.