VASOACTIVITY OF THE VENTRAL AORTA OF THE LAMPREY, *PETROMYZON MARINUS*, AND THE HAGFISH, *MYXINE GLUTINOSA*

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In recent years, we have discovered that a variety of receptors for vasoactive hormones and paracrines are expressed in the ventral aorta of the shark, *Squalus acanthias* (e.g., Evans, D.H., *J. Comp. Physiol.* 162: 179-183, 1992; Evans, D.H. et al., *J. Exp. Zool.* 265: 84-87, 1993; Evans, D.H. et al., *J. Comp. Physiol.* 165: 659-664, 1996), as well as the ventral aorta of the eel, *Anguilla rostrata* (Evans. D.H. and Gunderson, M.P., *Bull. MDIBL* 37: 107, 1998). Most recently, our data suggest that the endothelium-derived relaxing factor (EDRF) in the shark aorta is not nitric oxide (NO), but a prostaglandin (PGI_2; Evans, D.H. and Gunderson, M.P., *Am. J. Physiol.* 274: R1050-R1057, 1998). In fact, NO was actually constrictory in the shark. However, NO does dilate the ventral aorta of the eel (Evans, D.H. and Gunderson, M.P.., *Bull. MDIBL* 37: 107, 1998), suggesting significant differences in the action of this important molecule on the vascular smooth muscle of teleosts and elasmobranchs. This summer, we initiated a series of studies to extend these data to two agnathan species, in a effort to determine the evolution of the NO/PG system of EDRFs, as well as other vasoactive agonists. The lamprey and hagfish are the modern representatives of the earliest vertebrates, arising some 500 million years ago, but distinct from each other for most of that time, so they represent potential insights into the earliest evolution of these systems in the vertebrates.

Lampreys were collected in tributaries of Lake Erie and other Great Lakes by personnel of the U.S. Fish and Wildlife Service and Department of Fisheries and Oceans, Canada and shipped to MDIBL from the Hammond Bay Biological Station, Millersburg, MI. They were kept quarantined in well water in Living Streams maintained at 12 °C. Hagfish were purchased from the Huntsman Marine Laboratory, St. Andrews, NB, Canada, and maintained in running sea water at 16-18 °C. Lampreys were sacrificed by cervical section and double pithing; hagfish were anesthetized in 0.2% (V:V) 1-Phenoxy-2-Propanol. In both cases, the ventral aorta was removed and mounted as previously described (e.g., Evans and Gunderson, *Am. J. Physiol.* 274: R1050-R1057, 1998) in the appropriate Ringer's solution (12 °C) at 150-200 mg tension. No attempt was made to remove the endothelial lining. Putative constrictory and dilatory agents were added sequentially once the rings had stabilized (30-60 minutes after mounting). Data are expressed as mg change, mean±S.E. (N). All responses were significantly different from zero ($p < 0.05$) unless indicated.

Agonist	ET-1* 0.1 uM	ACh 0.1 mM	SNP 0.1 mM	NO 10 uM Initial	NO Final	pCNP 0.1 uM	Carb 1 uM	PGE1 1 uM
Lamprey	842±87 (18)	7.9±4.9 (13)	11.9±8.5 (9) (NS)	12.4±3.7 (14)	-110±25 (14)	-398±109 (9)	-111±32 (11)	- 884±124 (6)
Hagfish	105±22 (5)	ND	7.7±2.3 (5)	16.4±2.7 (5)	same	-83.6±17** (5)	-7.1±1.8 (5)	-11.2±4 (5)

*ET = endothelin; ACh = acetylcholine; SNP = sodium nitroprusside; NO = nitric oxide, pCNP = porcine C-type natriuretic peptide; Carb = carbaprostacyclin; PGE1 = prostaglandin E; **h-atrial natriuretic peptide

It is clear that the ventral aorta of both species express a variety of important receptors, each of which could play a major role in perfusion of the gills. ET-1 is a very potent constrictory agent in both species; interestingly, we found that sarafotoxin s6c was not very potent in the lamprey

(data not shown, and hag not tested), suggesting that the receptor in the lamprey is ET_A rather than ET_B, contrary to what we have described in the dogfish shark ventral aorta (Evans, D.H. et al., *J. Comp. Physiol.* 165: 659-664, 1996). ACh was not tested in the hagfish, but it is marginally constrictive in lamprey, contrary to what we described in the shark aorta (Evans, D.H. and Gunderson, M.P., *Experimental Biology On Line* 3: 3. 1998). The NO-donor, sodium nitroprusside produced little or no response in both species, but NO was constrictory in both species, with a biphasic response (a secondary dilation) in the lamprey. NO has never been suggested to be constrictory in vascular smooth muscle, so release of some secondary, constrictory substance may be involved. Porcine C-type natriuretic peptide was quite dilatory in the lamprey aorta, as was human ANP (data not shown). The hagfish aorta was also dilated by hANP; previously, we have shown that this tissue is sensitive to rat ANP (Evans, D.H., *J. Exp. Biol.* 157, 551-555, 1991) as well as porcine, killifish, and shark CNPs (Evans, D.H. et al. *Bull. MDIBL* 32, 106, 1993), so it appears that both species express both NPR-A and NPR-B receptors. Two prostanoids, carbaprostacyclin (a stable PGI_2 analog) and PGE_1 were dilatory in both species, as we have described for the shark ventral aorta (Evans, D.H. and Gunderson, M.P., *Am. J. Physiol.* 274: R1050-R1057, 1998). These data do not allow us to determine whether EP or IP receptors are involved in this response, since PGE_1 has been shown to interact with both EP and IP receptors, and carbaprostacyclin activates only the IP receptor (Coleman, R.A. et al., In: *Comprehensive Medicinal Chemistry*, eds. Hansch et al., 643-714, 1990). These data are the first physiological description of this array of vasoactive receptors in the vascular smooth muscle of two, distantly-related, agnathan species. (Supported by NSF IBN-9604824 and REU NSF BIR 9531348)