

Atlas of the Vasculature of Larval and Adult *Xenopus laevis*. Part: Interhyoideus Muscle

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In anuran larvae, the interhyoideus muscle provides the main force required to push water from the buccal cavity through the gill slits [1]. This movement is essential in feeding and respiration. In the obligate suspension feeding tadpoles of *Xenopus laevis*, the interhyoideus muscle is of considerable anatomical prominence. Albeit knowledge regarding the cross anatomical organization of the muscle's blood supply is well-established (see [2,3]), its microvascularization has not been subject to any study yet. In the present work, the microvascular anatomy of the interhyoideus muscle of *Xenopus* (from premetamorphic to juvenile stages) is surveyed. Data was gathered by scanning electron microscopy (SEM) of vascular corrosion casts and correlative light microscopy (LM) of paraplast-embedded stained tissue sections.

280 tadpoles of *Xenopus laevis* Daudin (stages 48 to 66 [4]) were used for vascular corrosion casting, 69 tadpoles for histology. Briefly, animals were killed by an overdose of an aqueous solution of tricaine-methane-sulfonate (MS 222; Sigma Chemicals, Basle, CH), the heart was exposed and the circulatory system was rinsed free of blood with Ringer solution via the arterial trunk with the venous sinus cut open to allow outflow of blood. After clear reflux from the opened sinus 0.5 ml Mercor-CI-2B, diluted 4+1 (v+v) with monomeric methacrylic acid containing 37.5 mg accelerator paste MA, were injected using a infuror pump (flow rate: 3-7 ml/h). After hardening of the injected resin tadpoles were macerated in 7.5% KOH (40°C), and vascular casts were cleaned, freeze-dried, mounted, evaporated with carbon and gold, and examined in the SEM (Cambridge 250, Cambridge, UK) at an accelerating voltage of 10 kV. For correlative light microscopy tadpoles were fixed by vascular perfusion with Bouin's solution. Specimens were embedded into paraplast, 7 µm thick transverse sections were made and stained with either hematoxyline–eosine or Goldner's trichrome stain.

Right and left external carotid arteries supply the respective portions of the interhyoideus muscle (IH) (Fig.1). Shortly before the IH the arteries bifurcate and their main branches split up into arterioles which finally capillarize on the dorsal surface of the IH (Figs. 2-4). In the paramedian areas arterioles form prominent networks of wide vessels. Occasionally, arterioles unilaterally connect via anastomoses, but also anastomose with contra-lateral partners crossing the median raphe separating right and left IH portions. Capillaries of the IH form a dense array of highly undulating vessels running parallel to the muscle fibers. Neighbouring capillaries frequently join via H-anastomoses. Postcapillary venules form small draining venules which merge beneath the surface of the IH to form larger veins that leave the muscle ventrally (Fig. 5). While intramuscularly, venules are dorso-ventrally flattened, but gain a more circular form upon emergence at the ventral surface

of the IH. Draining venules merge on the ventral surface of the IH to bilaterally form a ventral branch of the external jugular vein.

[1] N. Gradwell, *Herpetologica* **27** (1971), p107.

[2] N. Millard, *Trans. Roy. Soc. S. Africa* **30** (1945) p217.

[3] N. Millard, *Trans. Roy. Soc. S. Africa* **32** (1949) p55.

[4] P.D. Nieuwkopp P.D. and J. Faber, North Holland, Amsterdam, 1967.

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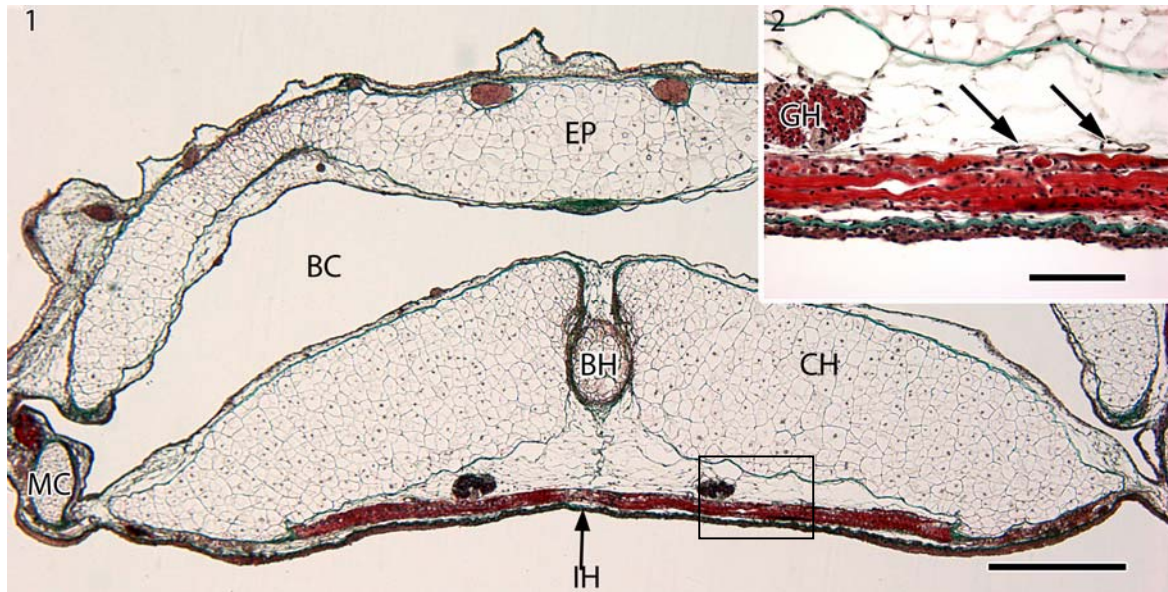


Figure 1. Histomorphology of the interhyoid muscle (IH) of larval *Xenopus laevis* at stage 60. Transverse section. 7 μm . Goldner stain. The muscle bilaterally inserts in the ceratohyale (CH), which is raised upon IH contraction. BC bucco-pharyngeal cavity, BH basihyale, EP ethmoidal plate, MC Meckelian cartilage. Scale bar: 500 μm . **Figure 2.** Close-up of the region indicated in Fig. 1 depicting two arterioles (arrows) paralleling the IH. GH geniohyoid muscle. Scale bar: 100 μm .

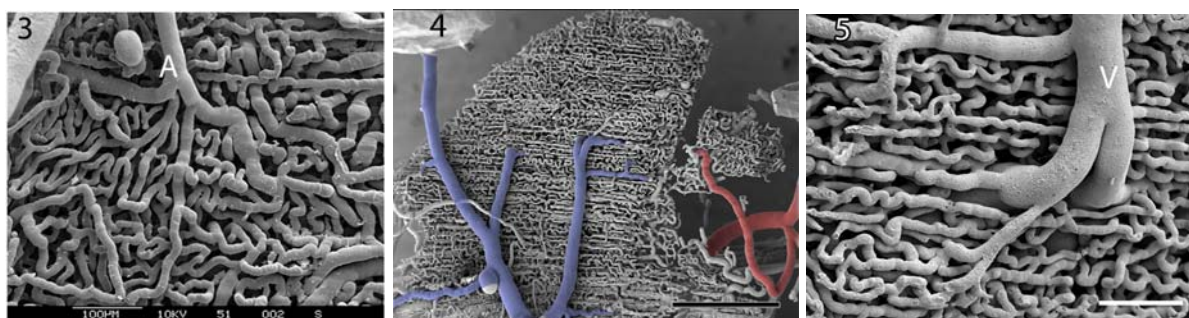


Figure 3 Arteriole (A) supplying the capillary bed of the dorsal aspect of the IH at larval stage 59. Vascular corrosion cast (VCC). **Figure 4.** Ventral aspect of IH at stage 60. VCC. Branches of the external carotid artery (in red) and of the external jugular vein (in blue) are seen. VCC. Scale bar: 500 μm . **Figure 5.** Venules emerging from the dense capillary bed of the IH at stage 60. VCC. Note the capillaries which outline the course of the muscle fibers. V venule. Scale bar: 100 μm .