THE ISOLATED PERFUSED GLOMERULUS OF THE ATLANTIC HAGFISH (MYXINE GLUTINOSA): A NEW PERFUSION TECHNIQUE.

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Hitherto, the glomerular filtration characteristics of Myxine were investigated in situ by perfusion of a 2.5 - 8 cm long portion of the eel-like body via the dorsal aorta. In such a preparation 6-15 glomeruli were perfused on each side of the body and urine was collected from both archinephric ducts (AND). Single nephron filtration rate (SNGFR) was obtained by dividing the urine flow rate by the number of perfused glomeruli. Using this technique valuable results were obtained on the protein permeability and the pressure dependence of glomerular filtration (Alt, et al., J. Exp. Biol. 91: 323-330, 1981; Rost, et al., Bull. MDIBL 23: 63-65, 1983).

To study basic filtration mechanisms of the glomerular barrier, the method was further developed. A double-barrelled perfusion cannula was inserted into the dorsal aorta. All arterial blood vessels were ligated except for the segmental artery and afferent arteriole leading to one glomerulus. The efferent arterioles were cut open to avoid that venous back pressure could influence the SNGFR (Alt et al., see above). Glomerular filtrate was collected via PP10 catheter which were distally introduced into the AND. SNGFR was directly obtained by the measurement of the advance of fluid in the catheter with a scale in the microscope. Variation of the perfusate flow permitted the regulation of the perfusion pressure which was measured in the dorsal aorta at the tip of the cannula. Pressure on the epithelial side of the glomerular capillary was obtained via another PP10 catheter proximal in the AND. This arrangement was isolated from all other tissue and transferred into a small chamber. Bathing medium (colloidfree Ringer's solution after Riegel, J. exp. Biol. 73: 261-277, 1978) was identical to the perfusate to avoid osmotic effects.

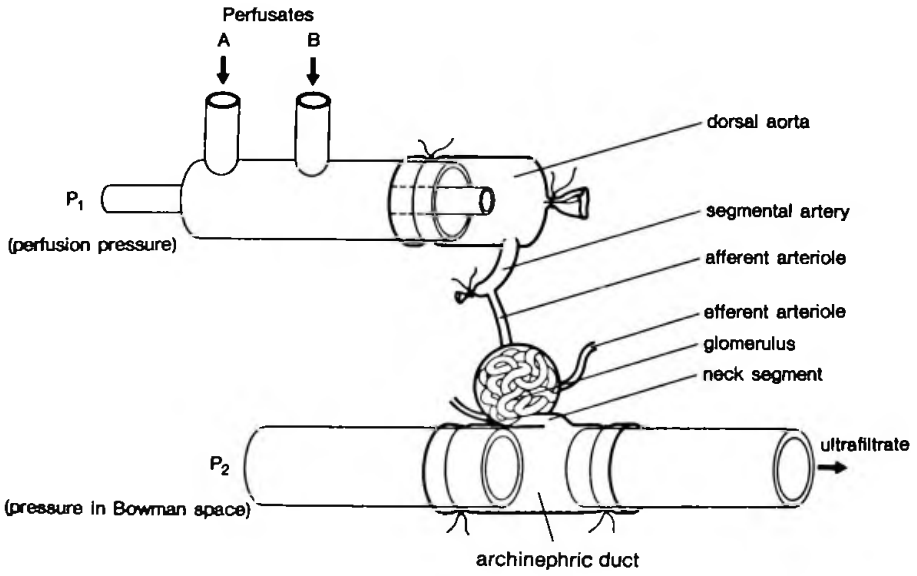

Fig. 1. In vitro perfusion of a single glomerulus of Myxine glutinosa

Advantages of the isolated perfused glomerulus:

1. The filtration fraction can be calculated on the basis of SNGFR and glomerular perfusate flow which equals the pump rate in this special preparation.

2. Simultaneous recording of perfusion pressure and pressure on the epithelial side of the glomerular capillary permits the calculation of the ultrafiltration pressure (P_{UF}).

3. The filtration area of every single glomerulus can be determined by morphometry as described by Aeikens (Virchows Arch. A. Path. Anat. and Histol. 381: 283-293, 1979). Pressure controlled perfusion fixation with Ringer's solution containing glutaraldehyde (1.25 g%) is easily performed by change of the perfusates.

4. The effect of hormones on the filtration dynamics can be studied without change of the perfusate and perfusate flow simply by adding the substances to the bath.

Preliminary results indicate that verapamil (Ca antagonist) has no significant effect in this preparation. SNGFR and P_{UF} were 153 nl/min and 9.4 cm H_2O, respectively (n=3). After the addition of verapamil (4.4×10^{-3} mol/l) either to the perfusate or the bath, SNGFR was 159 nl/min and P_{UF} was 9.7 cm H_2O. Therefore, it may be assumed that the blood vessels were maximally dilated under these experimental conditions.

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