

## THE CONTROL OF VENTRICULAR FLUID PRODUCTION IN THE DOGFISH

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Ventricular fluid (VF) production rate was determined in dogfish (*S. acanthias*) by perfusing an inulin containing VF-like buffer from the lateral ventricle to the cerebellum at a steady rate. By knowing the rate of inflow, and the inulin concentration in the inflow and the outflow, it is possible to calculate VF production rates. Each animal was given curare, and gills were perfused with running sea water. Needles were introduced stereotactically into one of the lateral ventricles and into the cerebellar ventricle. After satisfactory ventricular perfusion had been attained, a 2-3 hour control period was run. Ouabain or acetazolamide was then added to the perfusing fluid and VF production rates were determined for 4-6 hours. Several fish, instead, received intra-arterial ouabain. In 4 fish, using  $10^{-3}$  M ouabain, control production averaged 0.0060 ml/min, and a 63% decrease to 0.0022 ml/min occurred after addition of ouabain to the perfusate. In 3 fish concentration of ouabain in the perfusate was  $10^{-5}$  M; their average control production was 0.0070 ml/min, which decreased 44% to 0.0039 ml/min. In 2 fish using  $10^{-7}$  M ouabain in the perfusate no significant change in VF production (from 0.0041 to 0.0036 ml/min) was noted. In giving intra-arterial ouabain to dogfish it was noted that the approximate LD 50 for this drug was 0.1 mg/kg. Higher doses produced an initial increase in arterial pressure with increase in pulse pressure and heart rate, followed by slowing of heart rate, decrease in blood pressure and standstill of the heart in systole about 10-20 minutes after the administration of the drug. In 2 fish 0 mg/kg intra-arterial ouabain was tolerated well as determined by continuous arterial pressure monitoring. However, no change in VF production rate (from 0.0051 to 0.0052 ml/min) occurred. In 3 fish perfusing fluid containing 0.1 mg/ml of acetazolamide was used. This caused a 27% reduction in VF production rate (from 0.0056 to 0.0041 ml/min). This compares with a previously reported 50% reduction in VF production rate when acetazolamide is given intra-arterially at 3 mg/kg (Comp. Biochem. Physiol. 12, 171, 1964).

It thus appears that VF production rate in the dogfish is significantly reduced by high concentrations of intraventricular ouabain. Similar results may be obtained in the cat (Am. J. Physiol. 206, 1165, 1964). This reduction of VF formation rate may occur because of inhibition of Na-K activated ATP-ase by ouabain. It also appears that carbonic anhydrase is important in the regulation of VF production, as the strong inhibitor of the enzyme, acetazolamide, caused a significant reduction in VF production both when given into the ventricular system and into the bloodstream.

THE EFFECT OF FLUORIDE ION ON THE LONGEVITY OF *Campanularia flexuosa* HYDRANTHS

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Ageing might be considered according to Strehler a progressive loss of ability to adapt to

the environment. Lansing has suggested that ageing is the inability of protoplasm to maintain itself by self-synthesis. The sequence of events in the regression phase of the regression-replacement cycle of the colonial hydroid Campanularia flexuosa can be regarded as true ageing and death on a rigid cyclic program. Crowell has found that at a temperature of 17°C to 18°C the Campanularia lived from 4 to 9 days with the arithmetic average at 6.8 days. During a study of metabolic inhibitors on the life cycle it was observed that sodium fluoride appeared to effect longevity of the hydranths. Campanularia were maintained in sea water according to the methods of Crowell and the temperature of the water was maintained at 15-17°C. Sodium fluoride in concentrations ranging from  $10^{-3}$ M to  $10^{-5}$ M was added to the sea water culture containing the Campanularia. Preliminary statistical analysis suggests that sodium fluoride  $10^{-4}$ M increases the life span up to an arithmetic average of 8.3 days. The results of the Chi-square test substantially exceeded the 0.01 level of probability. The growth rate under these specific conditions increased by 2-3 fold. At a concentration of  $10^{-3}$ M the sodium fluoride decreased the life span to 6.1 days. The average life span for controls at 15°C was 6.75 days.

It is difficult to interpret this possible increase in longevity. It appears from the increased growth rate it is not a simple case of inhibition of metabolic rate. It is known that sodium fluoride at  $10^{-4}$ M will inhibit acid phosphatase activity, at  $10^{-2}$ - $10^{-3}$ M it will inhibit enolase, succinic dehydrogenase, myolinase, and enzymes containing Mg, Ca and other metals. Further investigation is in process to determine a possible enzyme target. The fact that sea water varies in many characteristics suggest caution in interpreting preliminary results using natural sea water as a solvent. Further work is in progress using larger sampling and artificial sea water.

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#### RESPIRATORY RATE AND SENESENCE IN Campanularia flexuosa

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The oxygen uptake of 100 - 200 Campanularia flexuosa hydranths at different stages in the regression-replacement cycle was measured using the Clark oxygen electrode and polarographic recording methods. Campanularia were cultured in sea water, using the technique of Crowell, and the temperature in the culture system and experimental vessels was maintained at 17-19°C. Respiratory rates of complete hydranths at four different ages were determined. There did not appear to be any significant differences in rate among complete hydranths 1, 2, 3 and 4 at the distal end. According to Crowell, position 1 is considered the youngest, and positions 3 and 4 the oldest. Preliminary determinations suggest that there may be a slight increase in respiratory rate at the onset of senility. This observation, however, needs further study since it is based on only 4 experiments utilizing only 500 individuals. The range of oxygen uptake for 100 hydranths in positions 1 - 4 was between  $10.5 \times 10^{-3} \mu\text{M O}_2/\text{min}/100$  hydranths to  $12.5 \times 10^{-3} \mu\text{M O}_2/\text{min}/100$  hydranths. Measurements of rate were determined for the first 15 minutes in the respiratory chamber. The data suggest that there is no decrease in respiratory rate during the complete hydranth stage. Regression appears to be a sudden climactic phenomenon, not preceded by a grad-