Table 2

GILL CLEARANCE OF MS 222 IN S. acanthias

Time min	Plasma μg/ml	Gill		Urine	
		Output μg	Clearance ml/min	Output μg	Clearance ml/min
0 - 5	30	9,000	60		
5 - 20	26	6,300	16		
20 - 35	22	3,200	10		
35 - 50	20	2,000	7		
50 - 170	16	5,400	3		
0 - 170 mean and cumulative values	20	26,000	8	10	.003

21 mg/kg injected at 0 time i.v. into 3 kg fish. The gill perfusion fluid (1.5 liters) was changed each period.

less due to compromise of physiological function including cardiac output, in the abnormal non-swimming environment (also applies to renal clearance, Table 1). The same effect was noted with all the drugs studied, although it was less for those drugs cleared slowly, i.e., less dependent on cardiac output. Fish were kept in the box up to eight hours; they became progressively anoxic, acidotic and lethargic. Most survived for several days when returned to the live-car in the sea, but generally did not recover normal activity.

The general conclusion is that the gill is relatively impermeable to drugs, compared with other membranes. Antipyrine for example enters the CSF, with a rate constant of 0.07 min^{-1} (Rall et al., J. Pharm. Exptl. Therap. 125:185, 1959), but from Tables 1 and 2 it may be judged that the corresponding value for gill exit (extrapolating to the free swimming fish) is about 0.003 min^{-1}. However, the relative gill clearance of drugs does reflect their lipid solubility, and at least one extremely lipid soluble compound, MS 222, is limited in its gill excretion only by gill blood flow.

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1966 #25

ACID-BASE PARAMETERS OF WILD VERSUS LABORATORY MAINTAINED DOGFISH—THE PROBLEM OF NORMAL VALUES IN MARINE BIOLOGY

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The demonstration that trawl captured dogfish, Squalus acanthias, maintained in captivity, have elevated arterial lactic acid concentrations (Bull. M.D.I.B.L., 5-2:30-31, 1965) prompted this study of acid-base parameters in wild fish. Dogfish were caught by handline in 5-10 fathoms of water, and blood samples were obtained within one minute. An 18 gauge needle was inserted into the caudal artery, 1 ml of blood was drawn in a non-heparinized syringe, then a heparinized

syringe was used for anaerobic sampling of another 10 ml blood sample for CO_2 content, pH, sodium, potassium, and chloride determinations. While the 10 ml sample was being drawn, a second person placed the 1 ml blood sample into 1 ml iced 10% perchloric acid for preparation of lactic acid filtrate. The filtrate prep..ration and the 10 ml anaerobically obtained sample were kept on ice. If blood sampling was not accomplished within one minute, the sample was discarded and the fish was not used. The boat returned to the laboratory 30 minutes after the first dogfish was caught. The dogfish were returned to the laboratory and maintained in live cars. Repeat arterial blood samples were obtained three to nine days after capture with the fish being maintained in a live car floated in the bay.

The data are presented in Table 1 and compared to similar data previously obtained in trawl-captured fish maintained in captivity.

Table 1

	Wild fish		Trawl captured fish
	Immediate	3-9 day capture	
pH	7.78 ± 0.07 (15)*	7.76 ± 0.06 (13)	7.52 ± 0.10 (17)
pCO_2 mmHg	3.07 ± 0.49 (15)	2.61 ± 0.62 (12)	2.9 ± 0.75 (17)
HCO_3^-†	7.15 ± 0.65 (15)	5.76 ± 1.02 (12)	3.9 ± 1.1 (17)
Lactate†	1.91 ± 0.63 (15)	4.08 ± 3.5 (13)	9.1 ± 5.3 (11)
Na^+†	252 ± 6 (14)	254 ± 15 (13)	263 ± 16 (9)
K^+†	3.44 ± 0.45 (14)	4.35 ± 0.56 (14)	4.1 ± 0.6 (10)
Cl^-†	240 ± 6 (14)	250 ± 10 (14)	249 ± 11 (9)
Hcrt.	20 ± 1 (7)	-	19 ± 3 (11)

*mean ± S.D. (number of specimens

†mEq/L

In the wild fish, there was not only the previously noted lower lactate concentrations, but arterial pH and HCO_3^- concentrations were higher than found in trawl captured fish. Arterial pCO_2 in wild fish was comparable to values found in the trawl captured fish. After the wild fish had been maintained in the live car for three to nine days, arterial blood lactic acid concentrations increased to above the wild state values, but were not as high as lactic acid of trawl captured fish. This increase in lactic acid concentration, while maintained in the live car, was accompanied by a decrease in arterial pCO_2 with maintenance of arterial pH at wild fish values.

It was of interest to note that the mean wild fish lactic acid concentration of 1.91 mM/L was higher than the 1.0 mM/L found in the 1965 series. Combining these two series for a total of 26 fish would yield a lactic acid concentration in wild fish of 1.52 ± 0.51 mM/L.

The trawl captured fish, when compared to wild fish, have decreased arterial blood pH presumably resulting from the lactic acid metabolic acidosis. The ability of the wild fish, when placed in the live car, to decrease pCO_2 and compensate for the increase in lactic acid thus maintaining a normal pH suggests that the marked elevation of lactic acid concentration in trawl

captured fish is not a function of respiratory restriction during captivity. Presumably, the marked lactic acidosis found in trawl captured dogfish is a result of prolonged struggle on the trawl coupled with the limited rate of lactic acid metabolism demonstrated in this species.

The potassium concentration in wild fish and trawl captured fish were not different, but there did occur an unexplained decrease in serum potassium in the wild fish maintained in captivity. The plasma chloride concentration of the wild fish increased 10 mEq/L during captivity.

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ACID PHOSPHATASE AND SENESCENCE IN Campanularia flexuosa

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The colonial hydroid Campanularia flexuosa is an excellent tool to study aging in that the events of the regression-replacement cycle represents true aging on a precisely cyclic time schedule. The relationship between acid phosphatase activity and this senescence cycle was investigated. Acid phosphatase levels were determined for Campanularia flexuosa hydranths of different ages. The colorimetric method used for the determination of acid phosphatase was the Sigma procedure utilizing Sigma 104 (p-nitro phenyl phosphate) phosphatase substrate.

The results were read at 400 mμ and 410 mμ using a Coleman Junior Spectrophotometer and semi-micro cuvettes. The data was expressed in Sigma units per mg protein. One Sigma unit of phosphatase will liberate 1 μM of p-nitrophenol per hour under the specified conditions of the technique (1 μM = 0.1391 mg). Approximately 135 hydranths of 5 different ages were compared through five series of experiments. The hydranths were homogenized in 1.25 ml of 5% Triton X-100. The Itzhaki and the Gornal Biuret techniques modified for small sample size were used to determine mg protein per ml. Complete hydranths in position 1, 2, 3, and 4 at the distal end of the hydrocaulus were compared with each other and with half stage seniles. Position 1 is considered the youngest hydranth and position 4 the oldest hydranth preceding the regression phase of senility. The mean values based on 4 series of experiments were compared.

Position	Acid phosphatase activity Sigma units/mg protein	Mean corrected acid phosphatase activity Sigma units/ml/135 hydranths
1	0.766	1.172
2	0.719	1.122
3	0.576	0.971
4	0.510	0.868
seniles	0.399	0.485

The results indicate that there is no increase in acid phosphatase concentration prior to the onset of senility. It appears that acid phosphatase is present in newly formed hydranths at peak levels and that senility involves a possible intracellular release and activation of acid phosphatase already present and associated with the lysozomes.

The results may further indicate a possible decrease in total acid phosphatase concentra-