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Uptake of Mercuric Chloride from Sea Water by *Serranus cabrilla*

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It is now well established that the major route of accumulation of mercury by fish is a direct uptake from water instead of a transfer through the food chain (HANNERZ 1968 , FAGERSTROM and ASELL 1973 , BOUQUEGNEAU and NOEL-LAMBOT 1977). This accumulation of mercurials in fish appears to be by way of the gills (OLSON et al. 1973 , BOUQUEGNEAU 1975).

Some recent studies about some sea water fishes reveal mercury uptake kinetics which can be described by the equation

$$C_t = C_{ss} (1 - e^{-Kt}) \qquad (1)$$

where C_t = concentration of mercury at time t

C_{ss} = asymptotic or steady-state concentration

$K = 0.693/t_{b1/2}$ being the theoretical biological half-time (PENTREATH 1975).

When considering the excretion of the pollutant, we report in table 1 the experimental half-times of elimination till now described in some species of sea water fishes. As it appears, they are relatively long and equal to about 100-200 days when considering inorganic mercury. When considering organic mercury, they are still greater, so that in the case of *Raja clavata*, no loss of mercury is detectable after 80 days in clean water and, in that case, the whole body accumulation can be described by a linear expression (PENTREATH 1976c).

When both kinetics of accumulation and release have been experimentally followed, it appears that the $t_{b1/2}$ calculated from the accumulation curve is more or less equal to the half-time of elimination of the pollutant observed when intoxicated fish are treated in clean water. Thus, for example, considering the kinetics of uptake of mercuric chloride by *Pleuronectes platessa*, PENTREATH (1976a) found a theoretical $t_{b1/2}$ of 187 days and, considering the slow and most important component of the kinetics of release, he found an elimination half-time of 162 days.

The generality of this rule is to be considered. Indeed, we describe and discuss in this paper the uptake kinetics of inorga-

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nic mercury by the Mediterranean teleost *Serranus cabrilla* which apparently presents a very short $t_{bl/2}$ calculated on the basis of the observed accumulation versus time curve and a much larger one when the release of the toxic material is measured in non-contaminated water.

TABLE 1

Biological half-times of mercury in some sea water fishes

Species	Pollutant	$t_{bl/2}$	References
Anguilla anguilla	CH_3Hg^+	1000 d.	JARVENPAA et al. 1970
Anguilla anguilla	$HgCl_2$	80 d.	BOUQUEGNEAU 1975
Pleuronectes platessa	$HgCl_2$	162 d.	PENTREATH 1976a
Pleuronectes platessa	CH_3HgCl	300 d.	PENTREATH 1976b
Raja clavata	$HgCl_2$	277 d.	PENTREATH 1976c
Raja clavata	CH_3HgCl	no loss detectable	PENTREATH 1976c
Serranus scriba	CH_3HgNO_3	267 d.	MIETTINEN et al. 1972

MATERIAL AND METHODS

Serranus cabrilla caught off Calvi's Bay (Corsica at S.T.A.R.E.S.O. (Station de Recherches Sous-Marines et Océanographiques de l'Université de Liège à Calvi) and weighing from 19 to 48 g were intoxicated in 80 l. aquaria containing aerated and unfiltered sea water to which 100 ppb mercury (as mercuric chloride) was added. Water was changed and polluted every day ; its temperature stayed close to 21°C. The fishes were not fed during the ten first days of intoxication.

To control the mercury concentration in the sea water during the same period, samples were filtered on a millipore filter (Type GS 0.22µ) in order to estimate the pollutant in solution and in suspension. Results are shown in fig.1 ; it appears that most of the mercury is dissolved in the sea water and, considering the loss of mercury in water resulting from the uptake by the fishes and described later, it appears that most of the mercury loss must result from evaporation and/or adsorption on the aquarium walls.

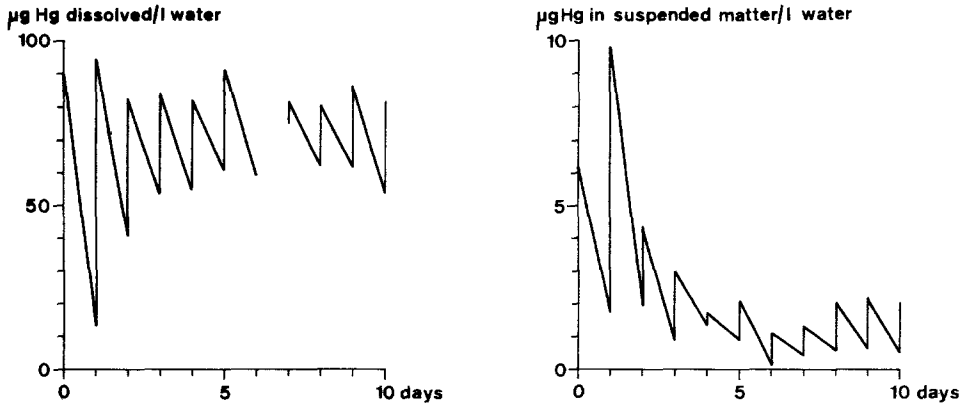

Fig.1 Concentrations of mercury in water during the ten first days of intoxication.

Four fishes were taken as samples before the experiment and after 1, 2, 4 and 10 days of intoxication. The remaining fishes were divided in two batches : the first continued to be intoxicated as before while the second was placed in 35 liters aquaria whose unpolluted and aerated water was changed every day and filtered continuously by means of activated charcoal and fiberglass filters in order to avoid any contamination of the sea water by mercury excreted by the animals.

The fishes were frozen and forwarded to the oceanological laboratory of Liège University where tissue samples were analysed for mercury by flameless atomic absorption spectrometry (Coleman Mercury Analyser System M.A.S. 50) after mineralization in H_2SO_4 95% and H_2O_2 30% as previously described (BOUQUEGNEAU 1973).

RESULTS

Table 2 shows the mercury concentrations in the four fishes analysed before intoxication. The values are rather high, probably owing to the well known mercury pollution level of the Mediterranean Sea.

Fig.2 shows the kinetics of uptake and release of mercury in the whole body during and after intoxication. All fishes (except one found dead on the ninth day) seemed to be healthy for the ten first days of intoxication. Those which were further intoxicated began to breathe quickly, to refuse feeding and finally died while the specimens which were put back in unpolluted water remained at first sight healthy and accepted feeding.

TABLE 2

Mercury concentrations (ppm ww) in *Serranus cabrilla* caught off Calvi's Bay

Organs	$\bar{x} \pm$ S.E.
Gills	0.23 ± 0.07
Liver	0.36 ± 0.15
Heart	0.85 ± 0.48
Digestive tract	0.18 ± 0.13
Gas bladder	1.05 ± 0.61
Kidneys	3.07 ± 1.86
Brain	0.64 ± 0.36
Head	0.17 ± 0.08
Muscle	0.18 ± 0.07
Gonad	0.18 ± 0.07
Whole body	0.19 ± 0.08

Each point of fig.2 represents the concentration of mercury in one fish from which the mean value of non-intoxicated fishes has been substracted so that the mercury concentration is put at zero before intoxication. The kinetic of accumulation has been adjusted to the equation $y = \alpha(1 - e^{-\beta x})$ by means of a programmed Hewlett Packard 9810 calculator, considering only the still living fishes. The calculated equation is

$$C_t = 1.7\ (1 - e^{-0.5t})$$

which shows a fast $t_{b1/2}$ equal to 1.36 day (0.693/0.5). This value is of course not in agreement with the experimental elimination half-time of about 100 days as suggested by the slow elimination of the pollutant from animals put back in clean water (dotted line of fig.2).

After eleven days of intoxication, the concentrations of mercury in fishes seem to be uncontrolled so that the observed data fit to a curve seemingly parallel to the initial rate of assimilation of mercury.

When considering individual organs (fig.3), accumulation curves present the same shape as the whole body. Concentrations at steady state, however, vary from 1 ppm for the muscles to 28 ppm for kidney, the other calculated values of C_{ss} being 6, 8 and 14 ppm for gills, digestive tract and liver respectively. In fishes found dead, a similar phenomenon of rapid uptake, as described in the whole body, appeared in each organ, leading to concentrations as high as 60, 210, 60 and 240 ppm, respectively, in gills, liver, digestive tract and kidney.

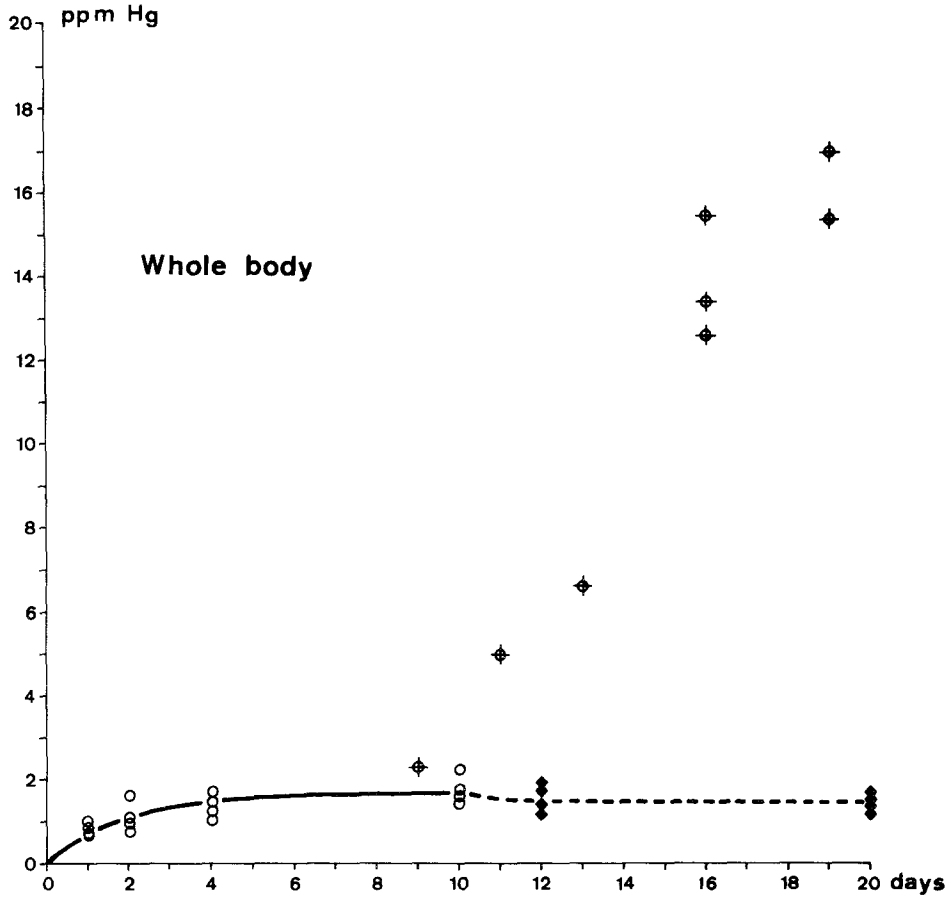

Fig.2 Kinetics of uptake (——) and release (- - -) of mercury in the whole body.

o intoxicated fish still alive
⊕ dead fishes
◆ intoxicated fishes put back in clean water

After ten days of elimination, only gills present an important and very significant loss of mercury (table 3). As far as it can be estimated with the two points presented in fig.3, the effective measured elimination half-time in the gills is about 5 days, which is, however, still greater than the calculated value of $t_{b1/2}$ from the accumulation kinetic (=0.7 days).

Table 3 further shows a relatively important loss of mercury in the digestive tract tissues. However, it appears that the loss of mercury from both gills and digestive tract does not account for the total loss of mercury from the whole animal. The

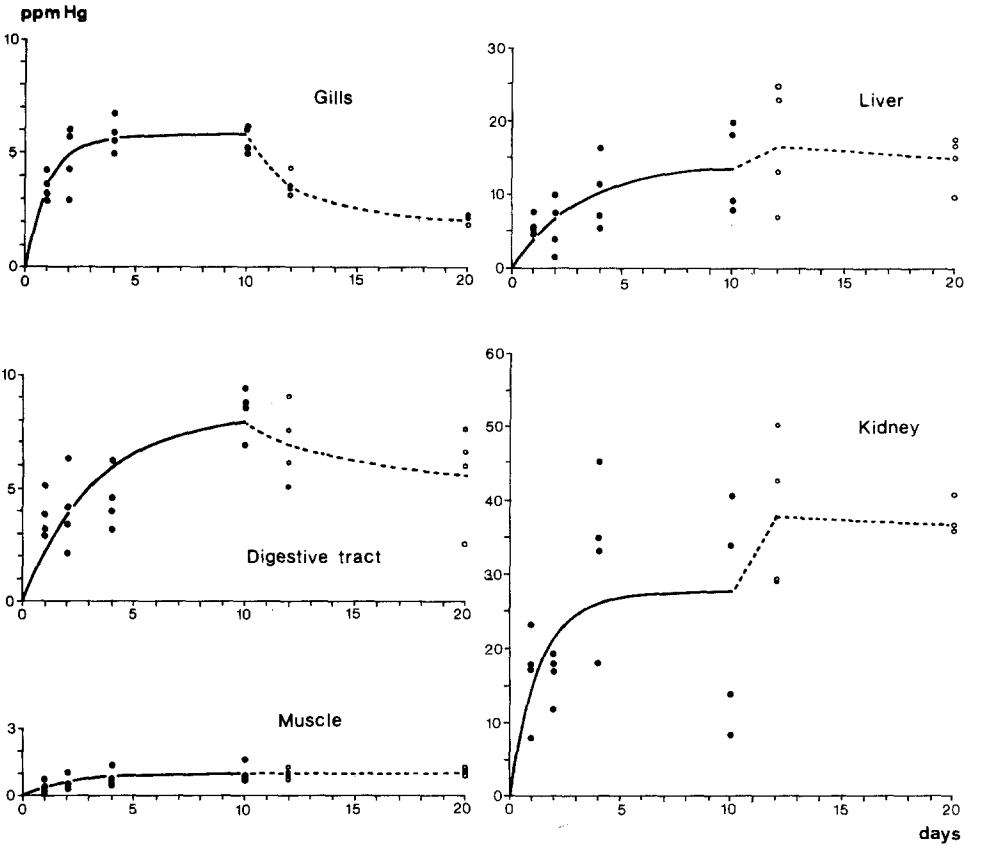

Fig.3 Kinetics of uptake (——) and release (----) of mercury in some organs of *Serranus cabrilla*.

difference results probably from an elimination in urine through the kidneys or/and in the bile by the liver. Both organs, however, present a slight increase of their mercury content after ten days in unpolluted water. Such an increase has been previously shown in other species of fish, for example by HANNERZ (1968) or by BOUQUEGNEAU (1975), and suggests a redistribution of the pollutant in the different organs of the animal towards the excretory organs.

TABLE 3

Comparison between the mercury concentration in fishes caught after ten days of intoxication and after identical treatment followed by exposure to clean sea water for ten days

Organs	ppm Hg (S.E.) in fish caught after 10 days of intoxication	+ 10 days of intoxication	t-test P	Δ µg/g organ	Δ µg/g fish
Gills	5.90 ± 0.31	2.32 ± 0.11	P < 0.001	2.58	0.11
Liver	14.33 ± 2.96	15.56 ± 1.87	0.8<P<0.9	-	-
Digestive tract	8.62 ± 0.53	5.85 ± 1.09	0.05<P<0.10	2.77	0.07
Kidney	27.45 ± 7.82	30.56 ±10.06	0.8<P<0.9	-	-
Muscles	1.20 ± 0.19	1.25 ± 0.08	0.8<P<0.9	-	-
Whole body	1.94 ± 0.17	1.66 ± 0.11	0.2<P<0.3	-	0.28

DISCUSSION

To summarize, it appears that the measured loss of mercury by *Serranus cabrilla* presents elimination half-times of the same magnitude as those previously described in other sea water fishes by several authors (see introduction). In the case of *Serranus cabrilla* however, the $t_{b1/2}$ calculated from kinetics described by the classical equation (1) is much shorter than expected from the mercury loss measurements. Such an observation can certainly not be attributed to a difference in the experimental protocols (relatively high concentration of 100 ppb in water compared to other experimental conditions, use of unlabelled mercuric chloride as pollutant, etc..) since, one of us (BOUQUEGNEAU 1975), using the sea water adapted eel *Anguilla anguilla* and the same technique has described uptake kinetics comparable to those described in *Pleuronectes platessa* and *Raja clavata* by PENTREATH (1976c). On the another hand, accumulation kinetics with short theoretical $t_{b1/2}$ have been described in fresh water fishes such as *Carassius auratus* (Mc KONE et al. 1971) and *Lebistes reticulata* (BOUQUEGNEAU and MERCENIER to be published).

Since such an accumulation kinetic does not reflect a rapid elimination of the pollutant from the animal, we are left to explain the apparent short half time of elimination by the induction by mercury itself of a decrease of the rate of entry of the pollutant. One mechanism which could be involved could, for example, result from an increase of the mucus production by the animal. Indeed, Mc KONE et al. (1971) have shown that mercury was concentrated initially in the external mucus secreted by *Carassius auratus* and that the presence of that pollutant in water appeared to

stimulate this secretion, some of which sloughed off.

In fact, we observed, but unhappily didn't quantify, an important mucus layer on gill's filaments of $HgCl_2$ intoxicated *Serranus cabrilla*. Further research will be carried out in our laboratory to test that hypothesis.

If, after some time in polluted water, such a mechanism was inhibited, the uptake kinetics would, rapidly, become quite linear in our conditions, leading to the death of the animals, for example by a breakdown of the osmoregulatory homeostasis as shown by BOUQUEGNEAU (1977) in the sea water adapted eel.

To conclude, it appears that important differences may exist between sea water fish species in their reaction to inorganic mercury exposure, leading to different concentration factors in similar environmental conditions depending on the relative efficiency of several entry control mechanisms, although elimination times remain of the same order of magnitude (100-200 days).

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