

Assimilation Efficiency of Dietary Methylmercury by Northern Pike (*Esox lucius*)

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Northern pike (*Esox lucius*) retained an average of 19% (range 6-31%) of the methylmercury which they ingested during consumption of young-of-the-year carp (*Cyprinus carpio*) collected from a pond; carp accumulated methylmercury naturally while in the pond. The total amount of mercury in pike increased with time (up to 42 d) but concentration in the tissue decreased due to growth dilution; duration of ingestion did not influence efficiency of methylmercury assimilation. This value (19%) is considerably lower than most efficiencies reported in the literature, demonstrating that methylmercury in this forage fish is less readily available to a predator fish than previous studies implied.

Key words: bioaccumulation, mercury compounds, predators, Esocidae

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Le grand brochet (*Esox lucius*) a retenu en moyenne 19% (variation de 6 à 31%) du méthylmercure ingéré après avoir consommé de jeunes carpes de l'année (*Cyprinus carpio*) tirées d'un étang. Durant leur séjour dans l'étang, les carpes concentraient naturellement le méthylmercure dans leurs tissus. La quantité totale de mercure présente dans le brochet a augmenté dans le temps (jusqu'à 42 jours), mais la teneur a diminué du fait de la croissance. La durée d'ingestion n'a pas influé sur le taux d'assimilation du méthylmercure. Ce taux (19%) est considérablement plus bas que la plupart de ceux qui sont signalés dans la documentation, ce qui montre que le méthylmercure accumulé dans la carpe n'est pas aussi facilement assimilable par un poisson prédateur que le laissaient croire les recherches antérieures.

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METHYLMERCURY has been extensively studied in aquatic environments, primarily because of the risks involved with human consumption of contaminated fish. Recently, workers in Sweden (Fagerstrom and Asell 1973; Fagerstrom et al. 1974) and in Canada (Nor-

strom et al. 1976) have attempted to model the accumulation of methylmercury by fishes; these models distinguish between methylmercury derived from water or from the diet. Although the models differ, a common feature is that the food component of methylmercury uptake is viewed as being proportional to the efficiency with which methylmercury is extracted from the diet.

Norstrom et al. (1976) have chosen a coefficient of 0.8 (80%) to describe dietary methylmercury assimilation based on the work of a variety of investigators (Hannerz 1968; de Freitas et al. 1974; Suzuki and Hatanaka 1975) while Fagerstrom and Asell (1973) have used a value of 0.15 (15%) after the work of Jernelov (1968). Because of the disparity of these co-

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efficients, this study was initiated to obtain more conclusive evidence on the efficiency with which a predator fish could assimilate methylmercury from a prey organism collected from nature.

Materials and methods — Northern pike (*Esox lucius*) obtained from a federal hatchery (U.S. Fish and Wildlife Service, Garrison Dam National Fish Hatchery, Riverdale, North Dakota) were fed young-of-the-year carp (*Cyprinus carpio*) collected from a pond located within the flood plain of the Tongue River, near Decker, Montana. Carp had accumulated methylmercury naturally while in the pond.

Pike were kept in four fiberglass living streams (Frigid Units, Inc., Toledo, Ohio) each of which was partitioned into four identical (400 L) compartments; two fish were housed in each compartment. Pike were fed rainbow trout (*Salmo gairdneri*) obtained from a local hatchery (U.S. Fish and Wildlife Service's Fish Cultural Development Center, Bozeman, Montana) during a 10-d acclimation period prior to the 42-d test. During the test pike were offered carp at an average rate of 9% wet weight/d. Pike were individually weighed and marked (fin clipped) at the onset; they were killed in groups of eight and subsequently analyzed for mercury after 13, 29, and 42 d. Surviving pike were reweighed at the killing intervals and their rations were readjusted to fixed percentages of body weight after each weighing. Eight pike were also killed for mercury analyses at the beginning of the test; the average mercury content of these ($0.10 \pm 0.01 \mu\text{g Hg/g}$) was assigned as the initial mercury content of each experimental pike. Eight more control pike (fed rainbow trout during the test) were killed and analyzed at the end.

Since pike were fed in groups of two, the amount of food (and therefore, the amount of mercury) fed to each individual was estimated as being proportional to the growth of the two fish. Thus, if a pair of pike were fed 100 g of food and their individual weight increases were 20 and 30 g, it was estimated that they consumed 40 and 60 g of food, respectively.

Groups of 10–15 carp were analyzed for total mercury 19 times during the test as were five groups of rainbow trout; results were used to calculate the amount of mercury fed to pike. This quantity was derived for fish killed after each interval from the total weight of carp fed and the average mercury content of carp analyzed up to that date. The mean mercury concentrations ($\mu\text{g Hg/g}$) in carp fed to pike over the three sacrifice intervals were 0.14, 0.12, and 0.10, respectively. Rainbow trout averaged $0.01 \mu\text{g Hg/g}$. Seven of the experimental pike and three groups of carp were also analyzed for methylmercury to determine the percentage of mercury present in this form.

Mercury concentrations in fish tissue were determined from aliquots of whole fish homogenates. Homogenates were prepared by blending whole pike (or groups of whole carp or trout) and a few grams of dry ice in a high speed blender. Blend samples were warmed to room temperature, resulting in a homogeneous paste.

Total mercury was determined by using a Varian model AA-6 atomic absorption spectrophotometer equipped with a carbon rod atomizer (Siemer and Woodruff 1974). Precision was estimated at $\pm 0.01 \mu\text{g Hg/g}$ based on the standard deviation of repeated analyses of the same sample.

Methylmercury was determined by homogenizing the fish, washing with acetone, freeing the methylmercury with hydrochloric acid, partitioning the methylmercury into

benzene, and finally quantifying using a Varian model GC-3700 gas-liquid chromatograph equipped with an electron capture detector (Watts et al. 1976). Error (determined from known standards) was relatively large ($\pm 30\%$) because the methylmercury concentrations analyzed were near the lower detection limit; this resulted in some percentages of methylmercury exceeding one hundred.

During the test, flow rates averaged 500 mL/min and photoperiod was 16 h light:8 h dark. Characteristics (mean) of the test water were: temperature 18.0°C , pH 8.31, dissolved oxygen 8.21 mg/L, total alkalinity (as CaCO_3) 69 mg/L, and hardness (as CaCO_3) 72 mg/L (APHA et al. 1976). Mercury was analyzed weekly in the test water but was never detected (lower detection limit $0.1 \mu\text{g Hg/L}$).

Results and discussion — Control pike, fed rainbow trout during the test, did not accumulate mercury. The mean mercury content of the trout was $0.01 \mu\text{g Hg/g}$ and pike that were fed trout averaged 55.2 g and $0.02 \mu\text{g Hg/g}$ after 42 d compared to 10.9 g and $0.10 \mu\text{g Hg/g}$ at the beginning. Therefore, the average control fish contained $1.10 \mu\text{g}$ of Hg at the end of the test compared to $1.09 \mu\text{g}$ of Hg at the onset. Thus, it is reasonable to assume that mercury accumulated by pike during the experiment originated from their diet (carp) and not from water. Norstrom et al. (1976) estimated fractional clearance of methylmercury at 30%/yr. This is less than 3.5% over 42 d; thus, we assumed that clearance was negligible during this test.

In fish analyzed for both total mercury and methylmercury, methylmercury was the predominant mercurial present. Methylmercury accounted for 88, 98, 103, 115, 90, 112, and 111% of the mercury in pike and 80, 115, and 124% of the mercury in carp. Other workers have reported similar findings (Bache et al. 1971; Westoo 1973). Based on these results, we assumed that all of the mercury in pike and carp was methylmercury.

The concentration of mercury in the experimental fish decreased throughout the test (due to growth dilution) but the total amount of mercury in pike increased with time (Table 1). Pike assimilated, on the average, 20, 20, and 17% of the mercury they consumed after 13, 29, and 42 d, respectively; these values are not significantly different, even at the 60% level of confidence (Student's *t*-test). The overall average during the experiment was 19%. This percentage is near that reported by Jernelov (1968), who fed "biologically synthesized" methylmercury to a predator fish, but lower than most values (38–89%) reported in the literature (Table 2). The important point here is that most workers have computed dietary methylmercury assimilation efficiencies from experiments during which methylmercury was offered to fish in a production diet (Lock 1975; Sharp et al. 1977; Phillips and Buhler 1978), orally administered in a water solution or tissue homogenate (Miettinen et al. 1970), or was present in a prey fish that had been exposed to high levels of methylmercury in water for a short duration (Jernelov 1968; Suzuki and Hatanaka 1975). Methylmercury added to a fish's diet by any of the above methods could conceivably be

TABLE 1. Growth, food consumption, mercury consumption, and mercury accumulation by northern pike fed young-of-the-year carp collected from the field. Values are means \pm 2 SE, ranges in parentheses.

Parameter measured	Sacrifice interval (d)			
	13	29	42	42 (control)
Pike weight (g)				
Initial	11.1 \pm 1.3 (9.5–15.2)	11.2 \pm 1.3 (9.9–14.9)	11.8 \pm 1.2 (9.9–14.4)	10.9 \pm 1.3 (9.7–14.6)
Final	17.0 \pm 2.3 (12.7–22.9)	32.9 \pm 3.3 (27.7–40.0)	55.7 \pm 7.2 (44.9–73.6)	55.2 \pm 6.7 (45.2–69.8)
Quantity of food fed (g) ^a	15.8 \pm 3.5 (8.0–21.2)	55.8 \pm 5.2 (46.6–66.0)	114.4 \pm 17.4 (86.0–152.2)	110.0 \pm 14.3 (92.5–153.1)
Mercury concentration in pike (μ g Hg/g)				
Initial ^b	0.10 \pm 0.01 (0.08–0.11)	0.10 \pm 0.01 (0.08–0.11)	0.10 \pm 0.01 (0.08–0.11)	0.10 \pm 0.01 (0.08–0.11)
Final	0.09 \pm 0.01 (0.08–0.12)	0.07 \pm 0.01 (0.05–0.09)	0.06 \pm 0.01 (0.03–0.08)	0.02 \pm 0.01 (0.01–0.03)
Quantity of mercury in pike (μ g Hg)				
Initial	1.12 \pm 0.13 (1.00–1.52)	1.12 \pm 0.13 (0.99–1.49)	1.18 \pm 0.12 (0.99–1.44)	1.09 \pm 0.09 (0.09–1.38)
Final	1.54 \pm 0.18 (1.02–1.83)	2.42 \pm 0.36 (1.66–3.19)	3.06 \pm 0.46 (1.93–4.03)	1.10 \pm 0.12 (0.95–1.48)
Change	0.43 \pm 0.14 (0.07–0.76)	1.30 \pm 0.33 (0.65–1.86)	1.87 \pm 0.36 (0.94–2.64)	0.01 \pm 0.07 (–0.21–0.17)
Quantity of mercury fed (μ g Hg)	2.21 \pm 0.49 (1.12–2.97)	6.69 \pm 0.63 (5.52–7.92)	11.44 \pm 1.74 (8.60–15.22)	— ^c
Mercury assimilated (%)	20 \pm 7 (6–31)	20 \pm 5 (9–29)	17 \pm 4 (7–28)	— ^c

^aCalculated as proportional to individual weight changes of pairs of northern pike that were kept in the same compartment.^bBased on the average mercury concentration of eight fish that were sacrificed at the onset.^cThese values were not calculated because both the change in the quantity of mercury in control pike and the concentration of mercury in the control diet were, for all practical purposes, zero.

loosely bound and therefore readily assimilated by a consumer fish, whereas methylmercury present in the diets of fish in nature may be associated with non-digestible components.

Archer et al. (1973) concluded that 80% of the methylmercury present in swordfish (*Xiphias gladius*) meat is bound to tissues resistant to digestion. Other metals, including strontium (Schiffman 1959) and zinc (Pentreath 1973) were reportedly less readily assimilated by fish from natural diets than from artificial diets. Thus, metals or metal derivatives such as methylmercury

are probably less available to fish in nature than many studies indicate. Future models of dietary methylmercury assimilation should employ a coefficient in the 15–20% range.

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TABLE 2. The efficiencies with which various species of fish were reported to have assimilated methylmercury from their diet.

Fish species	Diet used	Origin of CH ₃ -Hg ⁺ in diet	% CH ₃ -Hg ⁺ assimilated	Reference
Goldfish (<i>Carrassius auratus</i>)	Tetrafin goldfish food	Added during diet formulation	71–89	Sharp et al. (1977)
Rainbow trout (<i>Salmo gairdneri</i>)	Trout pellets (brand not specif.)	Added during diet formulation	52–71 ^a	Lock (1975)
Rainbow trout (<i>S. gairdneri</i>)	Oregon Test Diet	Added during diet formulation	68	Phillips and Buhler (1978)
Yellowtail (<i>Seriola quinqueradiata</i>)	Anchovy (<i>Engraulis japonica</i>)	Prey fish exposed in laboratory	67	Suzuki and Hatanaka (1975)
Northern pike (<i>Esox lucius</i>)	Orally administered water solution	Dissolved in solution	55	Miettinen et al. (1970)
Predator fish (not specified)	Prey fish (not specified)	Prey fish exposed in laboratory ^b	40–45	Jernelov (1968)
Northern pike (<i>E. lucius</i>)	Orally administered cow liver homogenate	Added during blending	38	Miettinen et al. (1970)
Predator fish (not specified)	Prey fish (not specified)	Prey fish collected ^b from nature	10–15	Jernelov (1968)
Northern pike (<i>E. lucius</i>)	Carp (<i>Cyprinus carpio</i>)	Prey fish collected from nature	19	This study

^aData for fish that consumed a diet containing 3.4 μ g Hg/g dry weight as methylmercury for 1–3 w.^bOrigin of CH₃-Hg⁺ in diet not actually stated but implied.

- AMERICAN PUBLIC HEALTH ASSOCIATION, AMERICAN WATER WORKS ASSOCIATION, AND WATER POLLUTION CONTROL FEDERATION. 1976. Standard methods for the examination of water and wastewater, 14th ed. American Public Health Assoc., New York, N.Y. 1193 p.
- ARCHER, M. C., B. R. STILLINGS, S. R. TANNENBAUM, AND D. I. C. WANG. 1973. Reduction in mercury content of fish protein concentrate by enzymatic digestion. *Agric. Food. Chem.* 21: 1116-1121.
- BACHE, C. A., W. H. GUTENMANN, AND D. V. LISK. 1971. Residues of total mercury and methylmercury salts in lake trout as a function of age. *Science* 172: 951-952.
- DE FREITAS, A. S. W., S. U. QADRI, AND B. E. CASE. 1974. Origins and fates of mercury compounds in fish, p. 31-36. *In* Proc. Int. Conf. on Transport of Persistent Chemicals in Aquatic Ecosystems, May 1-4, 1974, Ottawa, Ont.
- FAGERSTROM, T., AND B. ASELL. 1973. Methylmercury accumulation in an aquatic food chain. A model and some implications for research. *Ambio* 2(5): 164-171.
- FAGERSTROM, T., B. ASELL, AND A. JERNELOV. 1974. Model for accumulation of methylmercury in northern pike (*Esox lucius*). *Oikos* 25: 14-20.
- HANNERZ, L. 1968. Experimental investigations on the accumulation of mercury in water organisms. Fish. Board Swed. Inst. Freshwater Res. Drottningholm Rep. 48: 120-176.
- JERNELOV, A. 1968. Laboratory experiments on the change of mercury compounds from one into another. *Vatten* 24(4): 360-362.
- LOCK, R. A. C. 1975. Uptake of methylmercury by aquatic organisms from water and food, p. 61-79. *In* J. H. Koeman and J. J. T. W. A. Strik [ed.] Sublethal effects of toxic chemicals on aquatic animals. Elsevier Sci. Publ. Co., Netherlands.
- MIETTINEN, V., E. BLANKENSTEIN, K. RISSANEN, M. TILANDER, AND J. K. MIETTINEN. 1970. Preliminary study on the distribution and effects of two chemical forms of methylmercury in pike and rainbow trout. FAO technical conference on marine pollution and its effects on living resources and fishing. Rome, Italy. p. 1-12.
- NORSTROM, R. J., A. E. MCKINNON, AND A. S. W. DEFREITAS. 1976. A bioenergetic based model for pollutant accumulation by fish. Simulation of PCB and methylmercury residue levels in Ottawa River yellow perch (*Perca flavescens*). *J. Fish. Res. Board Can.* 33: 248-267.
- PENTREATH, R. J. 1973. The accumulation and retention of ⁶⁵Zn and ⁵⁴Mn by the plaice (*Pleuronectes platessa* L.). *J. Exp. Mar. Biol. Ecol.* 12: 1-18.
- PHILLIPS, G. R., AND D. R. BUHLER. 1978. The relative contributions of methylmercury from food or water to rainbow trout (*Salmo gairdneri*) in a controlled laboratory environment. *Trans. Am. Fish. Soc.* 107: 853-861.
- SCHIFFMAN, R. H. 1959. The uptake of strontium from diet and water by rainbow trout. *In* J. J. Davis [ed.] Hanford biology research annual report for 1958, HW-59500, Hanford Atomic Products Operation. Richland, Wash.
- SHARP, M. S., A. S. W. DEFREITAS, AND A. E. MCKINNON. 1977. The effect of body size on methylmercury clearance by goldfish (*Carassius auratus*). *Environ. Biol. Fish.* 2: 177-183.
- SIEMER, D. D., AND R. WOODRIFF. 1974. Application of the carbon rod atomizer to the determination of mercury in the gaseous products of oxygen combustion of solid samples. *Anal. Chem.* 46: 597-598.
- SUZUKI, T., AND M. HATANAKA. 1975. Experimental investigation on the biological concentration of mercury — II. On the origin of mercury found in the body of young yellowtail. *Bull. Japan. Soc. Sci. Fish.* 41: 225-231.
- WATTS, J. O. K., W. BOYER, A. CORTEZ, AND E. R. ELKINS JR. 1976. A simplified method for the gas-liquid chromatographic determination of methyl mercury. *J. Assoc. Off. Anal. Chem.* 59: 1226-1233.
- WESTOO, G. 1973. Methylmercury as a percentage of total mercury in flesh and viscera of salmon and sea trout of various ages. *Science* 181: 567-568.

