Archives of Environmental Contamination and Toxicology

# Mercury Accumulation in and Growth Rate of Rainbow Trout, Salmo gairdneri, Stocked in an Eastern Oregon Reservoir

Glenn R. Phillips<sup>1,2</sup> and Donald R. Buhler

Department of Agricultural Chemistry and Environmental Health Sciences Center, Oregon State University, Corvallis, Oregon 97331

Abstract. Mercury concentrations in lateral muscle tissue from rainbow trout ( $Salmo\ gairdneri$ ) stocked in a mercury contaminanted eastern Oregon reservoir increased linearly during the first five months that the fish were in the reservoir, followed by a leveling off period during which the mercury uptake curve became nearly asymptotic after eight months. In addition, the mean mercury concentrations present in three year classes of trout which had lived in the reservoir for 7, 19, and 31 months, respectively, were not significantly different, indicating that the uptake curve remains nearly asymptotic indefinitely. The shape of the curve is believed to be a result of exchange equilibria between water and tissue and factors that influence fish metabolism. It is estimated that 0.05  $\mu$ g Hg/L of methylmercury in water would have accounted for all of the methylmercury accumulated by trout in the reservoir. Growth rates (mg/g per day) of trout in the reservoir ranged from 0.7 in December to 39.7 in April, resulting in food consumption rate estimates ranging from 25 to 140 mg/g per day.

The finding in 1970 that fish from Lake St. Clair in southeastern Ontario contained elevated concentrations of mercury, triggered survey programs throughout the United States to examine the extent of mercury contamination in this country (Klein 1972). These surveys helped pinpoint areas of high mercury contamination and focused attention on some major sources of mercury to the environment (both natural and anthropomorphic). Included among the mercury problem areas implicated in the Pacific Northwest was Antelope Reservoir, an impoundment located about 70 miles south of Ontario, Oregon, near the Idaho border (Figure 1). Of 19 fish from this reservoir analyzed by Buhler *et al.* (1973), all but two had mercury concentrations in lateral muscle tissue exceeding the U.S. Food and Drug Administration's guideline level (0.5  $\mu$ g Hg/g) for human consumption.

<sup>&</sup>lt;sup>1</sup> Address correspondence to Glenn R. Phillips, Cooperative Fishery Research Unit, Montana State University, Bozeman, MT 59717

<sup>&</sup>lt;sup>2</sup> This research was conducted in partial fulfillment of the requirements for a Ph.D. degree in the Department of Fisheries and Wildlife, Oregon State University.

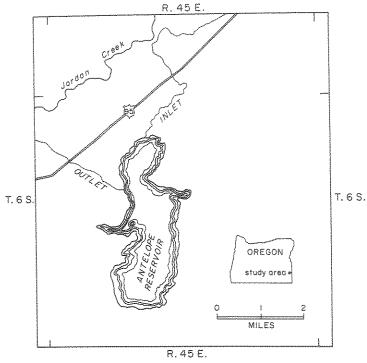


Fig. 1. Map of Antelope Reservoir showing the drainage ditches at the inlet and outlet. Inset map of Oregon shows location of study area

Mercury contamination in the reservoir is believed to have resulted from gold mining activities in the Jordan Creek drainage near Silver City, Idaho, during the 1860s. Using the "Washoe pan process," it was reported that nearly 2.5 tons of metallic mercury were lost into Jordan Creek between 1866 and 1869 (Gebhards et al. 1973). Hill et al. (1975) have reported concentrations of total mercury in sediments taken from Antelope Reservoir approaching 17 µg Hg/g.

The Oregon Department of Fish and Wildlife stocks fingerling rainbow trout (Salmo gairdneri) from their Oak Springs Hatchery (near Maupin, Oregon) in Antelope Reservoir each April. This stocking program provided an opportunity to sample fish which had lived in a mercury contaminated environment for known periods of time and to follow the growth of rainbow trout in a western reservoir.

### Materials and Methods

Fish were collected from Antelope Reservoir for total mercury analyses on 26 October 1972, 10 March 1973, 16 May 1973, 15 June 1974, 30 August 1974, 16 October 1974 and 16 December 1974, and from Oak Springs Hatchery near Maupin, Oregon, just prior to stocking of the reservoir on 14 April 1974; stocking typically occurs from early to mid-April in most years.

Fish were collected with a variable mesh gillnet; the net was set in late afternoon, fished overnight, and retrieved the following morning. The fish were weighed, measured, bagged, and

frozen for subsequent mercury analyses. Different year classes of rainbow trout in Antelope Reservoir were easily distinguished by size, since the reservoir provided an exceptional environment for growth.

Growth rates of the reservoir fish were calculated for monthly intervals by dividing the growth increment during each month by the mean fish biomass over the same interval. Monthly growth increments were estimated by interpolation between months when fish weights were actually measured. Food consumption rates were estimated from laboratory experiments during which rainbow trout were fed living tubificid worms (*Tubifex* sp.) at a series of fixed rates (Phillips 1976). The laboratory experiment was performed at 15°C, which approximates the average temperature of the reservoir during most of the growing season.

Prior to mercury analyses, a portion of lateral muscle tissue (5 to 10 g) was removed, weighed, and digested in nitric acid (3 hr) under reflux. The digest was oxidized with hydrogen peroxide and assayed by atomic absorption spectrophotometry (stannous chloride reductant, nitrogen carrier gas), using a Coleman Model 50 mercury analyzer equipped with a Soltec Model 52A recording unit (Magos 1971).

#### Results

Fish collected from the reservoir on 26 October 1972 included rainbow trout from three distinct year classes, representing residencies of 7, 19, and 31 months. No significant differences in mercury concentration existed between year classes (Table 1), which indicated that mercury concentration in muscle tissue must reach an equilibrium concentration sometime during the trout's first year in the reservoir.

Further studies were undertaken to investigate mercury uptake by trout during their first year in the reservoir. Fish stocked on 14 April 1974, were sampled at five intervals through December 1974. Total mercury concentrations in lateral muscle tissue increased steadily during the first five months the trout were in the reservoir, then leveled off and appeared to approach a steady state condition after eight months (Table 2 and Figure 2a). Further, the total quantity of mercury in trout declined as both the rates of fish growth (Fig. 2b) and mercury accumulation in tissue declined (Table 2 and Fig. 2c).

There is considerable evidence in the literature from both laboratory (Reinert et al. 1974; McKim et al. 1976) and field (Bache et al. 1971; Fagerstrom et al. 1974; Olsson 1976) investigations that support the notion that mercury uptake in fishes proceeds linearly for at least several months and often for several years. Accordingly, the average rate of mercury accumulation (micrograms of mercury accumulated per gram of fish per day) by trout in Antelope

Table 1. Mean total mercury concentrations in lateral muscle tissue from rainbow trout collected from Antelope Reservoir (26 October 1972) in relation to fish size and time spent in the reservoir

Sample size	Months in reservoir	Mean length ± SD (cm)	Mean weight ± SD (g)	Total Hg in muscle ± SD (µg Hg/g)
10	7	23.3 ± 1.4	145 ± 30	1.4 ± 0.3
10	19	$30.5 \pm 1.1$	$307 \pm 25$	$1.1 \pm 0.2$
10	31	37.3 ± 1.7	462 ± 71	$1.3 \pm 0.2$

Table 2. Actual or interpolated fish weights, mercury concentrations in muscle, and mercury body burdens for Antelope Reservoir rainbow trout following stocking on 14 April, 1974

Date <sup>a</sup>	Time in reservoir (days)	Mean weight <sup>h</sup> (g ± SD)	Mean mercury conc. in muscle <sup>c</sup> (μg Hg/g ± SD)	Estimated total mercury content of average fish <sup>(1)</sup> (µg of Hg)
April 14 (4)	0	9 ± 1	0.0 ± 0.0	0
May 16	32	42	0.2	8
June 15 (4)	62	$89 \pm 23$	$0.3 \pm 0.1$	27
July 15	92	170	0.5	85
Aug. 30 (3)	138	$462 \pm 152$	$0.7 \pm 0.1$	ę
Sept. 15	154	308	0.7	216
Oct. 16 (8)	185	$348 \pm 37$	$0.8 \pm 0.1$	278
Nov. 15	215	362	0.8	290
Dec. 16 (7)	246	$365 \pm 52$	$0.9 \pm 0.1$	329

a Values in parentheses are the actual number of fish sampled

Reservoir was calculated over the first five months of mercury exposure. Trout accumulated, on the average, 0.005  $\mu$ g Hg/g per day during this period.

Trout growth rates ranged from approximately 4% of body weight per day during April and gradually decreased to less than 0.1% by mid-December, while at the same time trout increased in mean weight from 9 to 365 g (Table 3). These growth rates correspond to food consumption rates ranging from 140 mg/g per day (milligrams of food consumed per gram of mean biomass per day) during late spring and early summer to 25 mg/g per day during mid-December (Table 3).

## Discussion

Rainbow trout in Antelope Reservoir accumulated mercury rapidly for several months, followed by a reduced rate of uptake. Several workers have studied mercury uptake by fish with respect to time in lentic environments. Fagerstrom and Asell (1973) observed a nearly asymptotic uptake curve for Swedish roach similar to the uptake curve seen for rainbow trout from Antelope Reservoir (Fig. 2a). Bache et al. (1971) sampling lake trout (Salvelinus namaycush) and Fagerstrom and Asell (1973) looking at northern pike (Esox lucius) observed more parabolic uptake curves. The curves for all of these species can be divided into three distinct phases: 1) an initial linear or almost linear phase during which mercury concentrations increase rapidly and nearly uniformly with re-

 $<sup>^{\</sup>rm b}$  Weights expressed as g  $\pm$  SD are actual mean weights for sampled fish and all other weights are interpolated from Fig. 2b

<sup>&</sup>lt;sup>c</sup> Mercury concentrations expressed as  $\mu g Hg/g \pm SD$  are actual concentrations for sampled fish and all other concentrations are interpolated from Fig. 2a

d Assuming muscle to be an average tissue with respect to mercury content

<sup>&</sup>quot;This value was not derived since only three fish were sampled on this date, two of which were unusually large

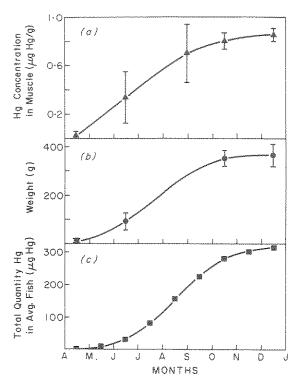


Fig. 2. Various trends observed in rainbow trout from Antelope Reservoir during 1974. The relationship between months spent in the reservoir, April (A) through January (J), and (a) mercury concentration in muscle ( $\mu$ g Hg/g); (b) weight (g), and (c) total quantity of mercury accumulated per fish ( $\mu$ g Hg). Each point represents the mean of from 3 to 8 fish  $\pm$  95% confidence intervals. All curves were fitted by inspection

spect to time; 2) a distinct levelling off phase when the rate of mercury accumulation rapidly decreases; and 3) a final equilibrium phase during which mercury concentration in the fish increases very slowly or remains relatively constant for several years.

The shapes of these mercury uptake curves are probably the result of several factors. Factors affecting the metabolic rates of fishes (e.g. temperature or dissolved oxygen concentration) have a large influence on the accumulation of mercury from water (Fagerstrom and Asell 1973) as do age-related decreases in metabolic rate (Brett 1965). However, in view of the fact that the third phase of mercury uptake was observed to be stable over several years (Table 1), a more encompassing factor is probably also responsible.

Hamelink et al. (1971) have explained DDT accumulation maxima in fish on the basis of exchange equilibria, and Ferguson et al. (1966) have similarly described endrin uptake. Apparently, these pesticides reach steady states between the animals and their environments governed by equilibrium conditions between water, blood and tissue. Assuming the same to be true for methylmercury, accumulation would initially proceed rapidly and uniformly, because of the large number of available binding sites; at some point in time, mercury accumulation rate would then begin to decrease, primarily due to the saturation of tissue binding sites, but also because of a reduced metabolic rate (hence reduced mercury intake via both food and water) associated with increased fish size. These phenomena probably account for the shapes of the mercury uptake curves observed for fishes in nature.

Table 3. Monthly estimates of growth rates and food consumption rates for Antelope Reservoir rainbow trout following stocking on 14 April, 1974

Date	Time in reservoir (days)	Interval (days)	Mean weight <sup>h</sup> (g ± SD)	Weight change (mg)	Average biocontent (g)	Growth rate (mg/g per day)	Estimated food consumption rated (mg/g per day)
April 14	9	2,	1 = 6	13 000	26	39.7	140
May 16	32	7 %	42	47.000	99	23.7	88
June 15	62	30	89 ± 23	81,000	130	20.8	7.0
July 15	75	\ <del></del>	0/-	80,000	210	12.3	50
Aug. 15	123	 ) (**	250"	58,000	279	6.7	40
Sept. 15	154	) 5.c.	308	40,000	328	3.0	35
Oct. 16	185	30	$348 \pm 37$	000.6	356	8.0	25
Nov. IS Dec. 15	215 246	, m	362 $365 \pm 52$	8,000	364	0.7	25

" Fish sampled on August 30 were not included in the derivation of growth rate, since only three fish were sampled on this date, two of which were unusually large

b Weights expressed as g ± SD are actual mean weights and all other weights are interpolated from Fig. 2b

d Estimates are derived from laboratory experiments (Phillips 1976) and are limited to nearest 5 mg/g per day "Mean fish weights for the indicated time interval

Adult rainbow trout sampled from Jordan Creek (Hill  $et\ al.$  1975) contained considerably less mercury (0.32  $\mu g\ Hg/g$  on the average) than trout from Antelope Reservoir that were analyzed during this study (Tables 1 and 2). Other workers have found a similar relationship between the concentrations of mercury found in fish from impoundments and their respective feeder streams (Gebhards  $et\ al.$  1973; Buhler  $et\ al.$  1973). Higher mercury concentrations in the reservoir trout are believed to result from Antelope Reservoir possessing a greater capacity for methylmercury formation than Jordan Creek. The water temperature, dissolved oxygen concentration, pH, and organic sediment load existing in the reservoir (Personal communication, Bill Hosford, Oregon Department of Fish and Wildlife) are similar to the conditions described as enhancing methylation of mercury by Jernelov (1972) and Fagerstrom and Jernelov (1972).

During this study, food consumption rate estimates for rainbow trout in Antelope Reservoir ranged from 140 mg/g per day (14%) in summer to 25 mg/g per day (2.5%) during late fall and winter. Similarly, Hakonson *et al.* (1975) reports values ranging from 8.2% in summer to 0.04% in winter, for rainbow trout in a Colorado bog lake. The decrease in winter is the combined result of reduced fish activity and decreased food availability associated with decreasing water temperature. The high rate of food consumption during summer reflects the abundance of food in Antelope Reservoir.

The methylmercury concentration in water that could have independently accounted for the observed mercury accumulation rate by fish from the reservoir (0.005  $\mu$ g Hg/g per day) can be estimated from the laboratory experiments of Phillips and Buhler (1978). Using this approach, 0.05  $\mu$ g Hg/L as methylmercury in water would account for all of the methylmercury accumulated. Similarly, brook trout (Salvelinus fontinalis) contained about 0.4  $\mu$ g Hg/g after three months (compared to 0.5  $\mu$ g Hg/g for rainbow trout after three months in Antelope Reservoir) exposure to 0.05  $\mu$ g/L of methylmercury in water (temperature 9-15°C) in experiments conducted at the National Water Quality Laboratory in Duluth, Minnesota (McKim et al. 1976); thus, the Duluth experiments lead to a similar prediction.

Detection limits reported for methylmercury in water ( $\mu$ g Hg/L) include 0.1 (Bisogni and Lawrence 1974), 0.05 (Stevens and Robertson 1974) and 0.004 (Chau and Saitoh 1973); however, most natural waters contain interfering substances that reduce detection limits. Klein *et al.* (1978), in their recent assessment of current analytical capabilities state, "It is not now possible to analyze methylmercury in water at the required levels." It is therefore unlikely that methylmercury hazard in natural waters can currently be evaluated by the analysis of water.

Although methylmercury concentrations in aquatic insects are generally low (Jernelov and Lann 1971; Cox et al. 1975; Hildebrand et al. 1976; Phillips 1976), concentrations of methylmercury present in fish from contaminated environments are consistently high. The implication is that dietary methylmercury would then be more important in piscivorous fish species than to insectivorous ones. A suggested hypothesis is that water accounts for a substantial amount of the mercury observed in both insectivorous fish species, such as trout in Antelope Reservoir, and in known piscivores. Fish eaters, such as

northern pike, acquire an additional increment of mercury through their diet. This hypothesis accounts for northern pike having consistently higher mercury concentrations than other fish species living in the same environment (Johnels et al. 1967; Jernelov and Lann 1971; Walter et al. 1974). The observation that squawfish (Buhler et al. 1973; Standiford et al. 1973) contain higher concentrations of mercury than coinhabiting species in some environments, but not in others, possibly reflects the degree to which this species depends on fish as opposed to insects for food in different environments.

Acknowledgments. We are indebted to the Oregon Department of Fish and Wildlife for their assistance in various aspects of this study. This work was supported by PHS Grants No. ES-00040, ES-00055 and ES-00210. Manuscript issued as Technical Paper No. 4805 from the Oregon Agricultural Experiment Station.

#### References

- Bache, C. A., W. H. Gutenmann, and D. V. Lisk: Residues of total mercury and methylmercury salts in lake trout as a function of age. Science 172, 951 (1971).
- Bisogni, J. J. Jr., and A. W. Lawrence: Determination of submicrogram quantities of monomethyl mercury in aquatic samples. Environ. Sci. Technol. 8, 850 (1974).
- Brett, J. R.: The relation of size to rate of oxygen consumption and sustained swimming speed of sockeye salmon (Oncorhynchus nerka). J. Fish. Res. Board Can. 22, 1491 (1965).
- Buhler, D. R., R. Claeys, and W. Shanks: Mercury in aquatic species from the Pacific Northwest. In D. R. Buhler (ed.): Mercury in the western environment, p. 59. Oregon State University, Corvallis: DCE Press (1973).
- Chau, Y. K., and H. Saitoh: Determination of methylmercury in lake water. Int. J. Environ. Anal. Chem. 3, 133 (1973).
- Cox, M. F., H. W. Holm, H. J. Kania, and R. L. Knight: Methylmercury and total mercury concentrations in selected stream biota. In D. D. Hemphill (ed.): Trace Substances in Environmental Health. p. 151. Vol. IX, University of Missouri: Columbia (1975).
- Fagerstrom, T., and B. Asell: Methylmercury accumulation in an aquatic food chain. A model and some implications for research. Ambio 2, 164 (1973).
- Fagerstrom, T., B. Asell, and A. Jernelov: Model for accumulation of methylmercury in northern pike (*Esox lucius*). Oikos 25, 14 (1974).
- Fagerstrom, T., and A. Jernelov: Some aspects of the quantitative ecology of mercury. Water Res. 6, 1193 (1972).
- Ferguson, D. E., J. L. Ludke, and C. G. Murphy. Dynamics of endrin uptake and release by resistant and susceptible strains of mosquito fish. Trans. Am. Fish. Soc. 95, 335 (1966).
- Gebhards, S., J. Cline, F. Shields, and L. Pearson: Mercury residues in Idaho fishes. In D. R. Buhler (ed.): Mercury in the western environment. p. 76. Oregon State University, Corvallis: DCE Press (1973).
- Hakonson, T. E., A. F. Gallegos, and F. W. Whicker: Cesium kinetics data for estimating food consumption rates of trout. Health Phys. 29, 301 (1975).
- Hamelink, J. L., R. C. Waybrant, and R. C. Ball: A proposal: Exchange equilibria control the degree chlorinated hydrocarbons are biologically magnified in lentic environments. Trans. Am. Fish. Soc. 100, 207 (1971).
- Hildebrand, S. G., A. W. Andren, and J. S. Huckabee: Distribution and bioaccumulation of mercury in biotic and abiotic compartments of a contaminated river-reservoir system. In R. W. Andrew, P. V. Hodson and D. E. Konasewich (ed.): Toxicity to biota of metal forms in natural waters. p. 211. Great Lakes Research Advisory Board, International Joint Commission, Windsor, Ontario (1976).

- Hill, S., A. Cochrane, D. Williams, M. Lucky, K. Greenfield, R. Farlee, B. Hudson, T. Tkachyk, D. Ugstad, P. Ugstad, and L. Wickman: Study of mercury and heavy metals pollutants in the Jordan Creek drainage. The Silver City Project, Student Originated Studies, National Science Foundation pub. 113, College of Mines, University of Idaho, Moscow, ID (1975).
- Jernelov, A: Factors in the transformation of mercury to methylmercury. In R. Hartung and B. D. Ginman (ed.): Environmental mercury contamination. p. 167. Ann Arbor, MI: Ann Arbor Sci. Pub. (1972).
- Jernelov, A., and H. Lann: Mercury accumulation in food chains. Oikos 22, 403 (1971).
  - Johnels, A. G., T. Westermark, W. Berg, P. I. Persson, and B. Sjonstrand: Pike (*Esox lucius L.*) and some aquatic organisims in Sweden as indicators of mercury contamination in the environment. Oikos 18, 323 (1967).
  - Klein, D. H.: Some general and analytical aspects of environmental mercury contamination. Environ. Chem. 49, 7 (1972).
  - Klein, D. H., A. Calabrese, F. M. D'Itrie, J. M. McKim, G. R. Phillips, D. J. Reish, and W. A. Willford: Mercury, In R. V. Thurston, R. C. Russo, C. M. Fetterolf, T. A. Edsall, and T. M. Barber Jr. (ed.): Review of the EPA Red Book: Quality criteria for water. pp. 20-1-20-5. Water Quality Section, American Fisheries Society, Bethesda, MD (1978).
  - Magos, L.: Selective atomic-absorption determination of inorganic mercury and methylmercury in undigested biological samples. Analyst 96, 847 (1971).
  - McKim, J. M., G. F. Olson, G. W. Holcombe, and E. P. Hunt: Long-term effects of methylmercuric chloride on three generations of brook trout (*Salvelinus fontinalis*): Toxicity, accumulation, distribution, and elimination. J. Fish. Res. Board Can. 33, 2726 (1976).
  - Olsson, M.: Mercury level as a function of size and age in northern pike, one and five years after the mercury ban in Sweden. Ambio 5, 73 (1976).
  - Phillips, G. R.: Some quantitative aspects of mercury accumulation by rainbow trout. Ph.D. Thesis. 129 pp. Oregon State University, Corvallis (1976).
  - Phillips, G. R., and D. R. Buhler: 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 (1978).
  - Reinert, R. E., L. V. Stone, and W. A. Willford: Effect of temperature on accumulation of methylmercuric chloride and p, p' DDT by rainbow trout (Salmo gairdneri). J. Fish. Res. Board Can. 31, 1649 (1974).
  - Standiford, D. R., L. D. Potter, and D. E. Kidd: Mercury in the Lake Powell ecosystem. National Science Foundation pub. Washington, DC (1973).
  - Stevens, A. A., and E. A. Robertson: Determination of methylmercury in water. Arch. Environ. Contam. Toxicol. 2, 266 (1974).
  - Walter, C. M., H. G. Brown, and C. O. Hensley: Distribution of total mercury in the fishes of Lake Oahe. Water Res. 8, 413 (1974).

Manuscript received August 28, 1978; accepted October 28, 1978.