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Uptake and Retention of Dietary Cadmium in Mallard Ducks

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Adult mallard ducks fed 0, 2, 20, or 200 ppm of cadmium chloride in the diet were sacrificed at 30-day intervals and tissues were analyzed for cadmium. No birds died during the study and body weights did not change. The liver and kidney accumulated the highest levels of cadmium. Tissue residues were significantly correlated in all treatment groups and residues increased with treatment level. Hematocrits and hemoglobin concentrations were normal in all groups throughout the study. Little cadmium accumulated in eggs of laying hens, but egg production was suppressed in the group fed 200 ppm.

INTRODUCTION

Cadmium is a toxic heavy metal that is rapidly becoming widespread in the environment. Its toxicity has been shown experimentally in several animal groups (Parizek, 1957; Sangalong and O'Halloran, 1973; Richardson *et al.*, 1974) and cadmium may produce histological changes in the kidney, liver, gastrointestinal tract, testis, heart, blood vessels, bone marrow, and pancreas. Cadmium also is known to induce testicular hypoplasia in birds and mammals and to inhibit spermatogenesis when fed or injected (Parizek, 1957; Richardson *et al.* 1974; White *et al.*, 1978a).

Little information is available on the uptake and distribution of cadmium in waterfowl exposed to cadmium through food chains. Cadmium has been detected in low to moderate amounts (<10 ppm) in kidneys and livers of canvasback ducks (*Aythya valisineria*) and ruddy ducks (*Oxyura jamaicensis*) (White and Kaiser, 1976; White *et al.*, 1978b), but it is not known whether those levels affect survival and reproduction. We conducted a chronic feeding study with mallards (*Anas platyrhynchos*), using levels of cadmium similar to or exceeding those that might be obtained by wild ducks through the food chain. We examined the uptake and retention of dietary cadmium in eggs, blood, brain, muscle, gonad, liver, and kidney at various intervals and evaluated the effects of cadmium on certain blood characteristics. Histopathological effects on kidneys and testes are reported elsewhere (White *et al.*, 1978a).

MATERIALS AND METHODS

Seventy pairs (one male, one female) of 1-year-old mallards obtained at the Patuxent Wildlife Research Center were randomly assigned to 90 × 90 × 60-cm vinyl wire cages supplied with running water. A long-day light cycle (14L:10D)

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was maintained throughout the study. After a 3-week equilibration period, the ducks were fed *ad libitum* with commercial duck breeder mash coated with either 2, 20, or 200 ppm (wet weight) of cadmium chloride dissolved in propylene glycol. Propylene glycol alone was mixed into the control diet. Three feed samples from each treatment group were taken at intervals throughout the study and frozen for cadmium analysis. Treatment groups fed 2, 20, and 200 ppm of cadmium consisted of 20 pairs each; the control group consisted of 10 pairs. Food intake per pair was estimated during the study.

After 30, 60, and 90 days on treated food, random samples of five pairs from each treatment group were weighed, bled, and sacrificed; controls were sacrificed after 30 and 90 days only. The remaining five pairs from each group were fed untreated food for 30 days and then sacrificed. All birds were necropsied and tissues were saved for cadmium analysis. All eggs laid during the treatment period were collected and one egg from each laying hen was randomly selected for cadmium analysis. A sample of the entire egg content (yolk and albumen) was analyzed.

The cadmium analyses were conducted at the Environmental Trace Substances Research Center, Columbia, Mo. A portion of each sample was ashed with a nitric and perchloric acid mixture, digested in an acidic solution, and assayed on a Perkin–Elmer Model 303 atomic absorption spectrophotometer. Limits of quantification were 0.005 ppm on a wet weight basis.

RESULTS

Mean levels of cadmium in the feed samples were 0.08, 1.6, 15.2, and 210 ppm (wet weight) for the control and 2-, 20-, and 200-ppm groups, respectively. The ducks consumed an average of 110 g of food daily and maintained body weights averaging 1153 g. Food consumption, body weights, and liver weights did not differ significantly ($P>0.05$, analysis of variance) among treatment groups throughout the study.

No birds died during the study. Necropsy showed that the birds were in good flesh and most contained fat deposits. Kidney weights in the 200-ppm group were significantly greater ($P<0.01$) after 60 and 90 days than those of controls, but testes in the 200-ppm group weighed less ($P<0.05$) after 90 days than those of controls (Table 1). Kidneys of birds fed diets containing 200 ppm of cadmium for 90 days and then fed untreated food for 30 days weighed significantly more ($P<0.01$) than those of controls. In addition, the testes of males in all treatment groups weighed less ($P<0.05$) than those of controls after 30 days post-treatment (Table 1).

There were no significant differences ($P>0.05$, analysis of variance) in hematocrits and hemoglobin concentrations between control birds and those fed treated food during the study. Average hematocrits were 47, 48, 45, and 45% and hemoglobin concentrations averaged 17, 15, 15, and 16 g/100 ml of whole blood for the control and 2-, 20-, and 200-ppm groups, respectively.

Average residue accumulations in the various tissues at each sacrifice interval were combined for males and females, since differences ($P>0.05$) were not detected between sexes. Residues increased in most tissues according to treatment level and time fed the cadmium diets (Table 2). Overall, residues in blood, brains,

TABLE 1
WEIGHTS OF KIDNEYS AND TESTES[a]

Cadmium treatment (ppm)	Interval (days) 30	60	90	30 (post-treatment)[b]
Kidney				
Control	2.72 ± 0.16	—[c]	3.63 ± 0.32	—[c]
2	2.74 ± 0.14	3.11 ± 0.22	3.77 ± 0.20	4.55 ± 0.63[d]
20	2.70 ± 0.11	3.33 ± 0.17	3.99 ± 0.29	3.19 ± 0.16
200	3.04 ± 0.18	4.01 ± 0.14[e]	5.73 ± 0.26[f]	4.68 ± 0.48[e]
Testis				
Control	7.53 ± 2.39	—[c]	7.31 ± 1.89	—[c]
2	10.05 ± 2.43	7.13 ± 2.73	9.31 ± 2.48	1.35 ± 0.59[d]
20	10.08 ± 2.10	6.35 ± 1.58	7.59 ± 2.83	2.96 ± 0.88[d]
200	8.26 ± 2.22	4.74 ± 1.78	2.39 ± 1.21[d]	3.69 ± 1.25[d]

[a] Organ weights (mean ± SE) are expressed in grams. N = 10 kidneys and 5 testes per treatment.
[b] Birds were fed treated food for 90 days and then untreated food for 30 days.
[c] Controls were not sacrificed at 60 days and at 30 days post-treatment; random samples from 30 days were selected for comparisons.
[d] Significantly different from controls, $P < 0.05$, analysis of variance.
[e] Significantly different from controls, $P < 0.01$, analysis of variance.
[f] Significantly different from controls, $P < 0.001$, analysis of variance.

[illegible] and gonads of birds in all treatment groups were low. Livers and kidneys [illegible]mulated the highest residues, especially those from birds in the 200-ppm [illegible]. Cadmium accumulations neared equilibrium in most tissues at about 60 [illegible] changing only slightly to moderately by 90 days (Table 2).

[illegible]sidues in tissues of birds fed cadmium for 90 days were similar to those of [illegible] of the post-treatment group (Table 2). In ducks fed clean food after cadmium [illegible]ment, residues declined ($P<0.01$) in blood of the 200-ppm group and increased [illegible]ains of the 20- and 200-ppm groups.

[illegible]ear regression analysis showed that significant correlations existed among [illegible] tissue combinations after 30 days of treatment. After 60 days of treatment, [illegible]es in all tissues were significantly correlated and after 90 days residue correla[illegible] were the highest, overall (Table 3).

Residues of cadmium in eggs differed significantly among treatment groups [illegible]ble 4), but egg laying was sporadic and the number of laying hens in each group [illegible] considerably. Residues in eggs within treatment groups were closely simi[illegible] (as indicated by the small standard errors), regardless of when the eggs were [illegible] (Table 4). Percentages of laying hens differed significantly among all treat[illegible] groups, but overall, the average number of eggs laid per laying hen in each [illegible] was small (controls = 3.5; 2 ppm = 3.3; 20 ppm = 3.7; 200 ppm = 1.0).

DISCUSSION

[illegible]etary or injected cadmium has been shown to accumulate mainly in the liver [illegible]idney of experimental animals (Cousins *et al.*, 1973; Dyer *et al.*, 1974; Pritzl [illegible] 1974; Horner and Smith, 1975), but its distribution varies according to its

TABLE 2
RESIDUES IN MALLARD DUCKS FED CADMIUM IN THEIR DIETS[a]

Cadmium treatment[b] (ppm)	Interval (days) 30	60	90	30 (post-treatment)[c]
Blood				
Control	0.022 ± 0.011	0.006 ± 0.000	0.003 ± 0.000[d]	—[e]
2	0.037 ± 0.022	0.009 ± 0.001	0.005 ± 0.001	0.003 ± 0.000[d]
20	0.029 ± 0.022	0.038 ± 0.003	0.068 ± 0.014	0.058 ± 0.036
200	0.096 ± 0.026	0.259 ± 0.019	0.330 ± 0.031	0.102 ± 0.011[f]
Brain				
Control	0.028 ± 0.019	—[e]	0.003 ± 0.000[d]	—[e]
2	0.015 ± 0.006	0.005 ± 0.001	0.003 ± 0.000[d]	0.004 ± 0.000
20	0.008 ± 0.002	0.010 ± 0.002	0.019 ± 0.002	0.051 ± 0.009[f]
200	0.040 ± 0.004	0.108 ± 0.005	0.116 ± 0.006	0.212 ± 0.020[f]
Muscle				
Control	0.006 ± 0.001	—[e]	0.015 ± 0.005	—[e]
2	0.012 ± 0.002	0.036 ± 0.026	0.016 ± 0.004	0.014 ± 0.002
20	0.025 ± 0.003	0.077 ± 0.033	0.082 ± 0.007	0.096 ± 0.016
200	0.147 ± 0.009	0.302 ± 0.016	0.501 ± 0.041	0.438 ± 0.010
Gonad				
Control	0.016 ± 0.005	—[e]	0.017 ± 0.008	—[e]
2	0.018 ± 0.007	0.157 ± 0.105	0.042 ± 0.014	0.455 ± 0.157
20	0.077 ± 0.017	0.248 ± 0.083	0.711 ± 0.243	1.653 ± 0.502
200	1.635 ± 0.994	3.220 ± 1.451	5.955 ± 1.964	11.443 ± 7.951
Liver				
Control	0.122 ± 0.016	—[e]	0.391 ± 0.107	—[e]
2	0.515 ± 0.079	1.344 ± 0.380	2.116 ± 0.375	2.732 ± 0.489
20	5.880 ± 0.744	14.400 ± 1.202	19.510 ± 3.426	19.130 ± 3.753
200	37.487 ± 6.249	109.630 ± 8.945	96.600 ± 5.702	81.900 ± 5.681
Kidney				
Control	0.547 ± 0.093	—[e]	0.459 ± 0.103	—[e]
2	1.537 ± 0.309	2.900 ± 0.613	4.627 ± 1.017	7.965 ± 1.726
20	13.390 ± 2.702	45.770 ± 4.500	54.333 ± 6.304	58.770 ± 8.499
200	88.130 ± 10.748	134.200 ± 4.744	77.222 ± 4.195	68.700 ± 6.508

[a] Residues in tissues are expressed in parts per million, wet weight (mean ± SE). N = 10 per treatment.
[b] Values for both sexes were combined since no differences existed between sexes.
[c] Birds were fed treated food for 90 days and then were fed clean food for 30 days.
[d] Trace values included in the calculations are half the limit of quantification (0.005 ppm).
[e] Controls not sacrificed at 60 days and at 30 days post-treatment.
[f] Residues at 30 days post-treatment significantly different ($P < 0.01$, analysis of variance) from those at 90 days.

existing form. Cadmium exposure induces the synthesis of metallothionein, a cadmium-binding protein, in the liver and possibly other organs (Cherian *et al.*, 1976). Free cadmium accumulates mainly in the liver, while metallothionein-bound cadmium is distributed primarily to the kidney and is characterized by high uptake and long retention (Tanaka *et al.*, 1975).

TABLE 3
CORRELATION AMONG RESIDUES IN MALLARDS FED DIETARY CADMIUM

Interval	Product-moment correlation coefficient					
	Blood	Brain	Muscle	Gonad	Liver	Kidney
30 days						
Blood	1	0.2249	0.4715[a]	0.2256	0.5216[a]	0.4965[a]
Brain		1	0.2361	0.4179[a]	0.3966[a]	0.3091[b]
Muscle			1	0.7028[a]	0.5396[a]	0.7836[a]
Gonad				1	0.7133[a]	0.6332[a]
Liver					1	0.7550[a]
60 days						
Blood	1	0.7415[a]	0.8087[a]	0.8591[a]	0.9827[a]	0.9017[a]
Brain		1	0.7925[a]	0.5918[a]	0.7177[a]	0.8162[a]
Muscle			1	0.6971[a]	0.8220[a]	0.7899[a]
Gonad				1	0.8626[a]	0.8119[a]
Liver					1	0.9030[a]
90 days						
Blood	1	0.9734[a]	0.7400[a]	0.9044[a]	0.9487[a]	0.9121[a]
Brain		1	0.9033[a]	0.9032[a]	0.9232[a]	0.8656[a]
Muscle			1	0.6761[a]	0.8487[a]	0.8396[a]
Gonad				1	0.9206[a]	0.8926[a]
Liver					1	0.9532[a]

[a] $P < 0.01$.
[b] $P < 0.05$.

Our results show that about 97% of the cadmium in mallard tissues was in the liver and kidney. After 90 days of treatment, birds in the 2- and 20-ppm groups accumulated residues in the liver similar to the concentrations in their diets and about 2.5 times that level in the kidney. In contrast, cadmium levels in livers and kidneys of the 200-ppm group averaged about half the dietary level. This relation suggests that the capacity of these organs to store cadmium is limited beyond a

TABLE 4
RESIDUES IN EGGS OF MALLARD HENS FED DIETARY CADMIUM

Cadmium treatment (ppm)	N[a,b]	Residue in eggs (ppm, wet weight)	
		Mean ± SE	Range
Control	4 (14)[c]	<0.007 ± 0.000	0
2	7 (23)[d]	<0.007 ± 0.000	0
20	15 (55)[e]	0.010 ± 0.001[f]	0.007 – 0.014
200	3 (3)[e]	0.040 ± 0.006[h]	0.031 – 0.051

[a] N = Number of laying hens in a treatment group; one egg per laying hen was analyzed for cadmium.
[b] The total number of eggs laid by the group of laying hens is indicated in parentheses.
[c,d,e] The percentages of laying hens per group were significantly different from one another ($P < 0.005$, chi-square test).
[f] Significantly different from the control and 2-ppm groups ($P < 0.05$, analysis of variance).
[h] Significantly different from the control and 2- and 20-ppm groups ($P < 0.001$, analysis of variance).

certain concentration. Cadmium concentrations in the kidneys of young swine did not increase substantially beyond a mean of 271 ppm, although the animals were fed diets of 450, 810, and 1350 ppm of cadmium for 6 weeks (Cousins *et al.*, 1973). Young Japanese quail (*Coturnix coturnix japonica*) accumulated about half their dietary level of 75 ppm of cadmium in the liver after 6 weeks of treatment (Richardson *et al.*, 1974).

Dietary cadmium did not cause significant body weight loss or decreased food consumption in our experimental mallards. Sturkie (1973) reported no significant weight loss in adult male chickens injected daily with 2 mg/kg of cadmium sulfate for up to 22 days. However, body weights of young chicks fed 75 ppm of cadmium decreased significantly after 3 weeks (Freeland and Cousins, 1973), as did the body weights of young Japanese quail fed 75 ppm for 6 weeks (Richardson *et al.*, 1974). Growth, as measured by weight gain, was significantly inhibited in young swine fed dietary cadmium (Cousins *et al.*, 1973), and dietary cadmium at 200 ppm for 23 days caused significant depression of body weight gain in growing male rats (Sansi and Pond, 1974).

Anemia is common in animals exposed to cadmium, regardless of the route of administration. Hematocrits were the most sensitive measure of toxicity in young swine fed cadmium (Cousins *et al.*, 1973). Male chickens injected with cadmium sulfate developed anemia and 34 days after treatment the hematocrits were still significantly depressed (Sturkie, 1973). Freeland and Cousins (1973) produced anemia in young chickens by feeding cadmium as did Richardson *et al.*, (1974) in young Japanese quail. In contrast, dietary cadmium did not significantly reduce hematocrits or hemoglobin concentrations in our experimental mallards. Since we used adult birds, this may be an age-related phenomenon, although anemia was produced in adult chickens after injection of cadmium (Sturkie, 1973).

In chickens, dietary cadmium at 60 ppm suppressed egg production (Sell, 1975), and 100 to 200 ppm eliminated egg production (Hennig *et al.*, 1968). Also, 600 ppm of dietary cadmium stopped egg laying in ducks, reduced hatchability in eggs laid before cessation, and impaired postnatal growth in ducklings (Hennig *et al.*, 1971). In our study, egg production appeared to be suppressed in hens fed 200 ppm of cadmium, but the control hens did not lay as many eggs as expected. In fact, a higher percentage of hens in the 2- and 20-ppm groups laid eggs than did the controls. We do not know the reason for the sporadic and highly variable egg production among hens in this study.

Our data agree with those from other studies (Sell, 1975; Hennig *et al.*, 1971) that have shown that little cadmium is transferred to the eggs of birds fed cadmium. Cadmium levels in eggs of our controls and the 2-ppm group were all <0.007 ppm, and, in eggs in the 20- and 200-ppm groups, levels averaged 0.01 and 0.04 ppm. Low levels of cadmium in eggs of wild birds, therefore, do not necessarily reflect low dietary intake of cadmium.

The results of this study should be useful in interpreting levels of cadmium found in wild ducks. An increase in dietary cadmium was reflected in an increase in tissue residues, suggesting a relationship between body levels and environmental contamination. Sharma and Shupe (1977) showed this in the natural habitat; cadmium accumulated in animal tissues as the environmental concentrations in-

creased. We found highly significant correlations among residue accumulations in the various mallard tissues analyzed. Our study however, did not include periods of weight gain or loss, which could alter these relationships. Further, our data showed that cadmium was retained at peak levels for at least a month in the liver and kidney but declined in the blood after a nontreated diet was fed for 30 days.

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REFERENCES

Cherian, M. G., Goyer, R. A., and Delaquerriere-Richardson, L. (1976). Cadmium–metallothionein-induced nephropathy. *Toxicol. Appl. Pharmacol.* **38**, 399–408.

Cousins, R. J., Barber, A. K., and Trout, J. R. (1973). Cadmium toxicity in growing swine. *J. Nutr.* **103**, 964–972.

Dyer, R. D., Born, G. S., and Kessler, W. V. (1974). The distribution of intraperitoneally injected cadmium-115 m in chickens *Environ. Lett.* **7**, 119–124.

Freeland, J. H., and Cousins, R. J. (1973). Effect of dietary cadmium on anemia, iron absorption, and cadmium binding protein in the chick. *Nutr. Rep. Int.* **8**, 337–347.

Hennig, A., Georgi, K., and Jeroch, H. (1971). Zur kontrazeptiven Wirkung des Kadmiums. *Arch. Exp. Vet. Med.* **25**, 793–798.

Hennig, A., Hartmann, G., and Anke, M. (1968). Kontrazeptive Wirkung von Cadmium bei Legehennen. *Naturwissenschaften* **55**, 551–552.

Horner, D. B., and Smith, J. C. (1975). The distribution of tracer doses of cadmium in the normal rat. *Arch Environ. Contam. Toxicol.* **3**, 307–318.

Parizek, J. (1957). The destructive effect of cadmium ion on testicular tissue and its prevention by zinc. *J. Endocrinol.* **15**, 56–63.

Pritzl, M. C., Lie, Y. H., Kienholz, E. W., and Whiteman, C. E. (1974). The effect of dietary cadmium on development of young chickens. *Poultry Sci.* **53**, 2026–2029.

Richardson, M. E., Spivey Fox, M. R., and Fry, B. E. (1974). Pathological changes produced in Japanese quail by ingestion of cadmium. *J. Nutr.* **104**, 323–338.

Sangalong, G. B., and O'Halloran, M. J. (1973). Adverse effects of cadmium on brook trout testis and on *in vitro* testicular androgen synthesis. *Biol. Reprod.* **9**, 394–403.

Sansi, K. A. O., and Pond, W. G. (1974). Pathology of dietary cadmium toxicity and the protective effect of injected iron. *Nutr. Rep. Int.* **9**, 407–414.

Sell, J. L. (1975). Cadmium and the laying hen: Apparent absorption, tissue distribution and virtual absence of transfer into eggs. *Poultry Sci.* **54**, 1674–1678.

Sharma, R. P., and Shupe, J. L. (1977). Lead, cadmium, and arsenic residues in animal tissues in relation to those in their surrounding habitat. *Sci. Total Environ.* **7**, 53–62.

Sturkie, P. D. (1973). Effects of cadmium on electrocardiogram, blood pressure, and hematocrit of chickens. *Avian Dis.* **17**, 106–110.

Tanaka, K., Sueda, K., Onosaka, S., and Okahara, K. (1975). Fate of ^{109}Cd-labeled metallothionein in rats. *Toxicol. Appl. Pharmacol.* **33**, 258–266.

White, D. H., Finley, M. T., and Ferrel, J. F. (1978a). Histopathological effects of dietary cadmium on kidneys and testes of mallard ducks. *J. Toxicol. Environ. Health.*, in press.

White, D. H., and Kaiser, T. E. (1976). Residues of organochlorines and heavy metals in ruddy ducks from the Delaware River, 1973. *Pestic. Monit. J.* **9**, 155–156.

White, D. H., Stendell, R. C., and Mulhern, B. M. (1978b). Relations of wintering canvasbacks to environmental pollutants—Chesapeake Bay, Maryland. *Wilson Bull.*, in press.