Heavy metal concentrations in the liver of two wild duck species: influence of species and gender

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ABSTRACT - The risk of wild ducks exposure to heavy metals in the environment was assessed by analyzing 20 wild ducks shot in the hunting area at the fish farm in Donji Miholjac, Eastern Croatia. Liver samples obtained from 10 Mallards and 10 Common Pochards were examined for heavy metals (Cd, Pb, As, Hg) by using flame atomic absorption spectrophotometry (AAS). Significant difference in heavy metal content between analyzed species was confirmed only for Cd (P ≤ 0.01), and no differences were found between genders within species. The results obtained suggest the importance of wild ducks as bioindicators of heavy metal pollution, especially Common Pochard for Cd, and Mallard for Pb accumulation.

Key words: Heavy metal, Mallard, Common Pochard, Eastern Croatia.

Introduction – Hunting is allowed at some freshwater fish farm sites in Eastern Croatia and these are important habitats for numerous waterfowl species. Mallard (Anas platyrhynchos) and Common Pochard (Aythya ferina) are very common in wetlands. Archaeologists discovered that in the Eastern Croatia (in Vučedol near Vukovar) people used meat of these ducks in 2.500 BC (Malez, 1995), and they are still using it today. Besides local hunters, the most common wild duck hunters arrive from Italy. Croatian Act on hunting prescribes that wild duck hunting must be carried out using a shotgun with shotgun pellets made mostly of lead. Consequently, the possibility of wild animals consuming lead shotgun pellets is real, as it was reported by Anderson et al. (2000). Also, if the available food on the water surface and at the fishpond bottom contains heavy metals, it endangers not only the fish, but also the fish-eating waterfowls. Since the wild duck meat is consumed by humans, the objective of this study was to examine the concentrations of four heavy metals (Cd, Pb, As, Hg) in liver of two wild duck species and to identify possible differences between species and/or gender.
**Material and methods** - As part of a broader survey on wild life exposure to heavy metals, ten Mallards (5 males, 5 females) and ten Common Pochards (5 males, 5 females) were collected in the hunting area at the fish farm in Donji Miholjac in Eastern Croatia. All birds were shot using lead shot during the hunting season in fall 2008 and individually put in labeled plastic bags. Liver samples were taken within 2 hours after death, weighted to 5g fresh weight and frozen in polypropylene containers at -20°C. The metal assays were carried out using flame atomic absorption spectrophotometry (AAS), according to Neugebauer et al. (2000). Statistical analyses were done using the analysis of variance, and the differences between means were analyzed using Fisher LSD. All statistical tests were performed using Statistica 7.0 software (StatSoft, 2004).

**Results and conclusions** – The results for heavy metal (Cd, Pb, As, Hg) contents found in liver tissue based on gender and species are shown in Table 1. It shows that concentration of heavy metals mainly does not exceed values prescribed by the Croatian law, except for lead and mercury based on population level. To determine whether the heavy metal concentrations found in the two species statistically differ, two variants of the ANOVA were introduced (Table 2). The first one is based on gender differences within species, and the second one on the differences between species. There are no significant differences between genders within species. The only significant difference between species is the cadmium content, which is highly significant (P≤0.01).

| Table 1. Heavy metal contents (mg/kg) found in liver tissues of Mallards and Common Pochard by AAS. |
|-----------------------------------------------|
|       | cadmium | lead | arsenic | mercury |
|       | male    | female | pop| male | female | pop| male | female | pop| male | female | pop| |
| Group 1 (Common Pochard)          |       |       |     |       |       |     |       |       |     |       |       |     |     |
| min | 0.016   | 0.001 | 0.001 | 0.001 | 0.001 | 0.040 | 0.046 | 0.040 | 0.001 | 0.001 | 0.001 | 0.001 | 0.001 |
| max | 0.280   | 0.115 | 0.280 | 1.621 | 3.517 | 3.517 | 0.078 | 0.081 | 0.081 | 0.016 | 0.277 | 0.277 | 0.001 |
| mean| 0.123   | 0.044 | 0.084 | 0.325 | 1.010 | 0.687 | 0.059 | 0.057 | 0.057 | 0.006 | 0.093 | 0.049 | 0.001 |
| ±SD | 0.121   | 0.046 | 0.096 | 0.724 | 1.525 | 1.182 | 0.017 | 0.014 | 0.015 | 0.006 | 0.115 | 0.089 | 0.001 |
| value1 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.100 | 0.100 | 0.100 | 0.100 |
| Group 2 (Mallard)               |       |       |     |       |       |     |       |       |     |       |       |     |     |
| min | 0.108   | 0.083 | 0.083 | 0.001 | 0.007 | 0.001 | 0.021 | 0.045 | 0.021 | 0.072 | 0.037 | 0.037 | 0.001 |
| max | 0.800   | 0.432 | 0.800 | 0.492 | 0.092 | 0.492 | 0.053 | 0.061 | 0.061 | 0.262 | 0.197 | 0.262 | 0.001 |
| mean| 0.418   | 0.249 | 0.332 | 0.158 | 0.061 | 0.113 | 0.040 | 0.052 | 0.046 | 0.122 | 0.111 | 0.116 | 0.001 |
| ±SD | 0.336   | 0.131 | 0.256 | 0.198 | 0.033 | 0.143 | 0.014 | 0.007 | 0.012 | 0.082 | 0.062 | 0.069 | 0.001 |
| value1 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.500 | 0.100 | 0.100 | 0.100 | 0.100 |

1 maximum allowable amount for internal organs in poultry by the Croatian law (Official Gazette, No. 16/2005); 2 pop=population.

Table 1 shows higher cadmium content found in Mallard. This difference is highly significant (P≤0.01) as it is presented in Table 2. The difference in Cd content can be explained by differences in feeding habit. Mallard are dabbling duck that feed mostly in agricultural
areas and at fishpond dyke. They feed on seeds and vegetative parts of aquatic and crop plants as well as on terrestrial and aquatic invertebrates (Kalisińska et al., 2004). Sampling place is located near a heavy traffic road, and cadmium is commonly found in oil derivatives, motor oils and tires (Gish and Christensen, 1973). Cadmium accumulation was reported in plants (Larison et al., 2000), which are the primary food for Mallard. Higher mercury content, which is not statistically significant, can also be explained in this migratory species in a similar way.

On the other hand, Common Pochard are diving duck which take herbal food mostly from water surface and animal food from water column and fishpond bottom. This explains that higher content of lead (Table 1) found in Common Pochard is likely caused by lead shotgun pellet consumed from the bottom of fishpond, and reabsorbed in gizzard (Clemens et al., 1975). This fact supports previous research which indicates that this species have highest prevalence of lead shot ingestion (Mateo et al., 2000). Our results suggest that Mallard is a good indicator of cadmium presence, and Common Pochard of lead accumulation, so these wild duck species can be used as important bioindicators of heavy metal pollution in the environment.

REFERENCES - Anderson, W.L., Havera, S.P., Zercher, B.W., 2000. Ingestion of lead and nontoxic shotgun pellets by ducks in the Mississipi Flyway. J. Wildl. Manage. 64, 848-857. Clemens, E.T., Krook, L., Aronson, A.L., Stevens, C.E., 1975. Pathogenesis of lead shot poisoning in the mallard duck. Cornell Vet. 65: 248-385. Gish, C.D., Christensen, R.E., 1973. Cadmium, nickel, lead and zinc in earthworms from roadside soil. Environ. Sci. Tehnol. 7(11): 1060-1062. Kalisińska, E., Salicki, W., Myslek, P., Kavetska, K.M., Jackowski, A., 2004. Using the Mallard to biomonitor heavy metal contamination of wetlands in northwestern Poland. Sci. of the Tot. Env. 320: 145-161. Larison, J.R., Linkens, G.E., Fitzpatrick, J.W., Crock, J.G., 2000. Cadmium toxicity among wildlife in the Colorado Rocky Mountains. Nature 406: 181-183. Malez, V., 1995. The findings of the bird remains of the Vučedol Site. Opvsc. Archeol. 19: 27-32. Mateo, R., Guitart, R., Green, A.J., 2000. Determinants of lead shot, rice, and grit ingestion in ducks and coots. Journ. of Wildlife Management 64: 939-947. Neugebauer, E., Sans Cartier, G.L., Wakeford, B.J., 2000. Methods for determination of metals in wildlife tissues using various atomic absorption spectrophotometry techniques, Tech. Rept. Ser No337E, Canadian Wildlife Service, Headquarters, Ottawa, 65p. Official Gazette No.16/2005 Ordinance on toxins, metals, metalloids and other harmful substances found in food. StatSoft, Inc. (2004): STATISTICA (data analysis software system), version 7. www.statsoft.com.

Table 2. Results of ANOVA based on gender and species as sources of variation.

| Gender as source of variation | DF | MS  | F      | P   |
|-------------------------------|----|-----|--------|-----|
| Cd                            | 18 | 0.05051 | 0.233153 | ns  |
| Pb                            | 18 | 0.77123 | 0.463479 | ns  |
| As                            | 18 | 0.00021 | 0.407800 | ns  |
| Hg                            | 18 | 0.00720 | 0.333565 | ns  |

Species as source of variation

| DF | Mean square | F      | P   |
|----|-------------|--------|-----|
| Cd | 18          | 0.3750 | 0.009969 | *  |
| Pb | 18          | 0.70866 | 0.155308 | ns  |
| As | 18          | 0.00018 | 0.062537 | ns  |
| Hg | 18          | 0.0635 | 0.076769 | ns  |

ns=not significant; *P≤0.01.