Health Risks associated with residual pesticide levels in fish reared in purified wastewater from slaughterhouse

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ABSTRACT. The main objective of the present research was to determine the concentrations of the selected pesticides in muscle, liver and skin of common carp. Fish were sampled in two different seasons from fish pond which received previously treated slaughterhouse wastewater. Pesticides including etridiazole, chloroneb, trifluralin, propachlor, chlorothalonil, hexa-chlorocyclopentadiene, atrazine, simazine, alachlor, metribuzin, metolachlor, DCPA, cyanazine, chlorobenzilate, endrin aldehyde, cis permethrin and trans permethrin were determined by using a GS-MS method. Many of pesticides were not determined or determined in low concentrations. Propachlor was found in muscle, skin and liver. The recommended acceptable daily intake was higher in comparison with the estimated daily intake for examined pesticides via fish reared in treated slaughterhouse wastewater. It is very important to maintain the safety of the fresh fish produced in wastewater in order to ensure food safety and avoid health problems in humans.

Keywords: common carp; food safety; environmental protection; integrated aquaculture system, propachlor, risk assessment

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INTRODUCTION

Fish meat is highly acceptable protein source worldwide. Moreover, fish represents an excellent source of fatty acids in healthy nutrition (Ljubojević et al., 2015). From another point of view, consumers are more and more oriented to the safety issues due to the potential presence of different environmental contaminants in fish meat. Consumption of fish contaminated with pesticides may lead to poisoning of consumers (Aamir et al., 2016). Pesticides are widely used in agriculture (Hladik et al., 2016). It is highly important to understand the nature, bioaccumulation and distribution of pesticides in aquatic environment and also the routes of contamination in order to control and prevent outbreaks of poisoning associated with fish consumption. Pesticides are present in aquatic environment and can be transferred into fish and enter into the food chain (Guo et al., 2008). Due to the fact that pesticides are highly soluble in fats, they accumulate in fish tissue (El-Shahawi et al., 2010). According to Cao et al. (2007) pesticides can remain in soil and water for years. There is increasing trend in the use of wastewater in fish production, particularly in developing countries. The fish rearing in waste water may represents a significant risk for public health (Kim and Aga, 2007). Fish may represent a bioindicator of environmental contamination and the health risk arising from fish produced in wastewater filled ponds is questionable. Thus, the safety of fish reared in wastewater filled ponds could be a serious concern for public health. There are several studies regarding the level of pesticides in common carp, mainly organochlorine pesticides in different parts of the world (Svobodová et al., 2003; Hu et al., 2010; Ondarza et al., 2010; Pelić et al., 2019), but no available data exists about the level of other pesticides in fish produced in treated slaughterhouse wastewater.

Data analysis was performed using Excel (Microsoft Excel, 2007) to determine the descriptive statistic parameters.
Table 1. Method performance data obtained in blank common carp samples, spiked with 50 µg/kg (n=20)

| Pesticides                  | LOD (µg/kg) | LOQ (µg/kg) | Precision (%) | Linearity (r²) | Recovery (%) | RSD (%) |
|-----------------------------|-------------|-------------|---------------|----------------|--------------|---------|
| Etridiazole                 | 1.5         | 4.9         | 4.8           | 1.00           | 97.4         | 6.2     |
| Chloroneb                   | 0.9         | 2.1         | 7.8           | 1.00           | 98.1         | 7.9     |
| Trifluralin                 | 0.4         | 1.3         | 0.8           | 1.00           | 99.3         | 9.2     |
| Propachlor                  | 0.3         | 1           | 9             | 1.00           | 99.6         | 8.3     |
| Chlorothalonil              | 0.4         | 1.2         | 5.4           | 1.00           | 98.1         | 12.6    |
| Hexa-chlorocyclopentadiene  | 1.2         | 4.1         | 3.4           | 1.00           | 97.3         | 3.8     |
| Atrazine                    | 0.4         | 1.2         | 4.4           | 1.00           | 90.4         | 8.1     |
| Simazine                    | 0.5         | 1.6         | 3.2           | 1.00           | 94.2         | 4.2     |
| Alachlor                    | 1.1         | 3.6         | 4.5           | 1.00           | 91.8         | 8.6     |
| Metribuzin                  | 0.8         | 2.6         | 8.3           | 1.00           | 89.3         | 8.9     |
| Metolachlor                 | 1.2         | 4.4         | 3.1           | 1.00           | 96.6         | 3.7     |
| DCPA                        | 1.3         | 4.8         | 3.8           | 1.00           | 95.4         | 3.9     |
| Cyanazine                   | 0.8         | 2.9         | 8.1           | 1.00           | 93.4         | 11.2    |
| Chlorobenzilate             | 1.1         | 3.7         | 4.7           | 1.00           | 91.6         | 10.3    |
| Endrin aldehyde             | 1.4         | 4.8         | 3.5           | 1.00           | 95.2         | 8.3     |
| Cis permethrin              | 1.2         | 4.2         | 3.8           | 1.00           | 96.4         | 3.4     |
| Trans permethrin            | 1.4         | 4.7         | 11.7          | 1.00           | 112.3        | 12.3    |

LOD, Limits of detection; LOQ, Limits of quantification; RSD, relative standard deviation

Table 2. Concentrations (µg/kg) of pesticides in meat, skin and liver of common carp (n = 7) in spring

| Pesticide          | Muscle tissue | Skin | Liver |
|--------------------|---------------|------|-------|
|                    | Frequency (%) | Range | X±SD  | Frequency (%) | Range | X±SD  | Frequency (%) | Range | X±SD |
| Propachlor         | 100           | 11.4-11.5 | 11.4±0.05 | 100          | 20.40-21.32 | 20.83±0.38 | 100          | 11.3-11.5 | 11.4±0.1 |
| Atrazine           | 14.29         | <lod-26.88 | / | 0              | <lod | / | 0              | <lod | / |
| Endrin aldehyde    | 71.43         | <lod-38.25 | 37.30±0.53 | 0              | <lod | / | 0              | <lod | / |

* All other analysed pesticides were below LOD.

Table 3. Concentrations (µg/kg) of pesticides in meat, skin and liver of common carp (n = 7) in autumn

| OC Pesticide        | Muscle tissue | Skin | Liver |
|---------------------|---------------|------|-------|
|                     | Frequency (%) | Range | X±SD  | Frequency (%) | Range | X±SD  | Frequency (%) | Range | X±SD |
| Chloroneb           | 0             | <lod | / | 28.57         | <lod-22.31 | 13.40±12.61 | 0              | <lod | / |
| Propachlor          | 100           | 20.46-20.59 | 20.53±0.06 | 100          | 20.46-34.15 | 24.49±6.59 | 100          | 20.47-20.74 | 20.58±0.12 |
| Atrazine            | 0             | <lod | / | 28.57         | <lod-115.83 | 102.51±18.84 | 0              | <lod | / |
| Simazine            | 14.29         | <lod-45.28 | / | 28.57         | <lod-91.99 | 85.27±9.51 | 0              | <lod | / |
| Alachlor            | 0             | <lod | / | 42.86         | <lod-90.78 | 78.53±21.13 | 0              | <lod | / |
| Metribuzin          | 0             | <lod | / | 28.57         | <lod-126.74 | 126.67±0.1  | 0              | <lod | / |
| Metolachlor         | 0             | <lod | / | 28.57         | <lod-44.09 | 44.11±0.2  | 0              | <lod | / |
| DCPA                | 0             | <lod | / | 14.29         | <lod-28.47 | 0          | 0              | <lod | / |
| Cyanazine           | 0             | <lod | / | 28.57         | <lod-404.85 | 385.8±26.92 | 0              | <lod | / |
| Chlorobenzilate     | 0             | <lod | / | 14.29         | <lod-138.31 | 0          | 0              | <lod | / |
| Endrin aldehyde     | 0             | <lod | / | 14.29         | <lod-111.13 | / | 0              | <lod | / |

* All other analysed pesticides were below LOD.
RESULTS

Table 1 shows method performance data for 17 selected pesticides. The results of the study are shown in Table 2 (spring) and Table 3 (autumn). Generally, the concentrations of pesticides were low in all analyzed samples in both seasons. A number of pesticides were not detected neither in muscle tissue, and neither in liver and skin samples. It includes the following: etridiazole, trifluralin, chlorothalonil, hexa- chlorocyclopentadiene, cis permethrin and trans permethrin. In autumn concentrations of propachlor were two fold higher compared to concentrations obtained in spring and were 20.53 ± 0.06 µg/kg in muscle tissue, in skin concentrations were in range 20.46 - 34.15 µg/kg and in liver was 20.58 ± 0.12 µg/kg. Atrazine was above the limit of detection in two samples of skin and simazine in one sample of muscle tissue and two samples of skin. Alachlor was found in three samples of skin and metribuzin in two. Metachlor, DCPA and cyanazine were found only in skin samples. Chlorobenzilate was found only in one sample of skin, while endrin aldehyde was present only in one sample of skin in amount of 111.13 µg/kg.

DISCUSSION

Concentrations of pesticides were generally comparable or lower in comparison with previous results reported in studies of common carp and Cyprinidae species worldwide (Svobodová et al., 2003; Darko et al., 2008). Etridiazole, chlorothalonil, atrazine, simazine, metribuzin, metolachlor, DCPA and chlorobenzilate are registered for use in Serbia. All of those pesticides except etridiazole and chlorothalonil were detected in low levels in skin of common carp. Pesticides such as atrazine, simazine, trifluralin, alachlor are listed as priority substances of the Directive 2008/105/EC (EEC, 2008) and the Directive 2013/39/EC (EEC, 2013) and three of those pesticides were detected in the present study. A number of pesticides not registered (chloroneb) or banned in Serbia were also found (mainly propachlor, alachlor and endrin aldehyde) and their presence could be attributed to illegal use. The pesticides reported in this study are not often examined so the data regarding their content in fish including common carp are scarce. Papadakis et al. (2015) reported the detection of etridiazole, metolachlor, atrazine, and alachlor (maximum concentrations of 0.026, 0.688, 1.465 and 1.098 µg/L, respectively, in the surface waters of Lake Vistonis Basin, Greece. Kong et al. (2008) highlighted that pesticides were used widely in China, and that pesticides like atrazine, simazine, and alachlor have a harmful effects on endocrine system. All the mentioned pesticides were detected but only in skin and in relatively low concentrations in common carp in our study. Inadequate regulation and poor agriculture management are the main reasons for the fact that banned pesticides are still in illegal usage. Among pesticides determined in the common carp samples of muscle, liver and skin residual concentration of propachlor was dominant in all samples in both season. The determined concentrations were the lowest in liver, then in muscle, while the highest levels were measured in skin. Ondarza et al. (2010) also reported that the pesticide levels were higher in muscle then in liver. Since the skin is a kind of protective barrier, it may be the reason for the highest concentrations of pesticides detected in it. Thomas et al. (2012) examined levels of pesticides in muscles of farmed common carp in France and also found that the levels of pesticides were low. Also, Padula et al. (2008) didn’t detect any pesticide residues in wild and farmed southern bluefin tuna (Thunnus maccoyii). Kong et al. (2008) evaluated the municipal sewage treatment system in China for their pollutant removal efficiency. They found that concentrations of alachlor, acetochlor, atrazine were 0.074 - 0.021 µg/l, 0.160 - 0.096 µg/l and 0.238 - 0.184 µg/l, respectively, and the total removal efficiency of atrazine was poorest through the sewage treatment systems. Our results showed that the wastewater treatment systems in slaughterhouse were efficient in removing pesticides, and consequently that the discharged effluent is not harmful for the aquatic environment. Furthermore, Khalil and Hussein (1997) reported that the primary and secondary treated waste effluents were successfully used to grow the Nile tilapia. The presented results showed that common carp could be successfully reared in treated slaughterhouse wastewater with regard to pesticides contamination. Janković et al. (2012) reported that estimated weekly intake of common carp in Serbia is 29.4 g. The risk related to pesticides via consumption of examined fish for human of body weight of 70 kg is compared with acceptable daily intake (ADI) of selected pesticides recommended by various organization (Table 4). The estimated daily intake (EDI) was significantly lower than ADI for each pesticide, even in the worst case scenario. Having that in mind, it can be concluded that this intake would not pose a health hazard in human populations.
CONCLUSIONS

Pesticide levels in all tissues of examined fish were below the permitted levels. The pesticides included in the present study are not usually included in the monitoring studies in Serbia but also in different states worldwide. However, the obtained results revealed that several of those pesticides could pose a significant human health hazard and also an environmental hazard. This should be taken into account by the authority so that those pesticides should be included in some new monitoring schemes. The knowledge and proper management of pesticides associated with the fish consumption are of considerable medical, economic and environmental importance. Continuous monitoring, including appropriate testing of fish meat in order to determine the presence of pesticides is necessary. It is important due to the fact that fish is an important food source but also an important indicator of environmental pollution. The obtained results provide important data for the risk assessment of pesticides from fish reared in integrated systems. However, the further research are necessary for developing risk management tools for integrated aquaculture production systems.

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CONFLICT OF INTEREST

On behalf of all authors, the corresponding author states that there is no conflict of interest.
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