Evaluation of Residues of D.D.T and D.D.A in Fish Collected from Caspian Sea, Iran

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ABSTRACT

Background: Pesticides are essential in modern agricultural practices but due to their biocide activity and potential risk to the consumer, the control of pesticide residues in foods is a growing source of concern for the general population. Extensive application of such agents as organochlorine pesticides in farmlands and contemporary agricultural industries has led to undesired environmental contamination and human health hazards. Thus, this study attempted to evaluate and analyze the residual values of the organochlorine insecticide D.D.T and its metabolite D.D.A in the four species of most consumed fish collected from the Caspian Sea.

Methods: In this investigation, concentrations of residual values of D.D.T and D.D.A were quantitatively determined in the 4 species of fish sampled from 4 major fishing centers (Chalous and Babolsar cities and Khazar Abad and Miankaleh regions) in Mazandaran province, Iran, using gas chromatography electron-capture detection (GC–ECD) in 2008.

Results: The results showed that the highest values of D.D.T were in Mugil auratns (0.033±0.008 mg/kg) and Rutilus frisikutum (0.031±0.007 mg/kg) fishes collected from Babolsar sampling center.

Conclusion: Concentrations of D.D.T and D.D.A in the fish were found to be less than the standard permissible intake.

Keywords: Caspian Sea, D.D.A, D.D.T, Fish, Organochlorine Pesticide.

INTRODUCTION

Food production capacity is faced with an ever-growing number of challenges, including a world population expected to grow and a falling ratio of arable land to population. The explosive increase in world population is mostly observed in developing countries and this is where the need for food is greatest and starvation threatens human life. Therefore, pesticides are essential in modern agricultural practices but due to their biocide activity and potential risk to the consumer, the control of pesticide residues in foods is a growing source of concern for the general population (1-3). On the other hand, environmental pollution and its hazards are the most important problems of societies and living creatures while increased population and the development of technology and production can cause a lack of attention to environmental safety. Industrialization leads to the pollution of ecosystems. Therefore, recognition of pollutants and prevention of their environmental dispersion are one of the necessities in this field which require

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determining the pollution sources, their environmental effects, and prevention methods as well as filtration of industrial waste water and education of instructions for environmental protection which play a vital role in control of and protection against pollutants (4-6).

Pollution is discharged into rivers and lakes and leaches into the soil and ground water, or is emitted into air as particulate matter (7-9). This has increased concerns about the accumulation of metals in sediments, biota, and ultimately humans (10,11). Fish, as human food, are considered as a good source of protein, polyunsaturated fatty acids (particularly omega-3 fatty acids), calcium, zinc, and iron (12). In the future, seafood will even be a more important source of food protein than they are today and the safety for human consumption of products from aquaculture is of public health interest (13). Fish from estuaries and coastal waters associated with industrial and sewage discharges have been found to be contaminated with pesticides (8,10,14).

Synthetic organochlorines, such as D.D.T, are highly resistant to degradation by biological, photochemical, or chemical means. They are liable to bioaccumulation, toxic, and probably hazardous to human and/or environmental health and most of them are prone to long-range transport. These compounds are also typically characterized by having low water solubility and high lipid solubility (4). Organochlorines have been associated with significant environmental impact in a wide range of species and at virtually all tropic levels. They have also been implicated in a broad range of adverse human health and environmental effects, including impaired reproduction, endocrine disruption, immunosuppression, and cancers. Exposure to organochlorines has been correlated with population decline in a number of marine mammals (15,16).

The primary transport routes into marine and coastal environments are atmospheric deposition and surface runoff, the former being by far the greatest albeit dispersed over large areas. Because many organochlorines are relatively volatile, their re-mobilization and long-distance redistribution through atmospheric pathways often complicate the identification of specific sources (4).

The Caspian Sea, the largest inland sea in the world, is bordered by five countries: Iran, Azerbaijan, Turkmenistan, Kazakhstan, and Russia. It has no outlets and acts as a reservoir for water in the region. Environmental pollutants found in the sea probably arrive via the Mazandaran and Gilan rivers. Industrial complexes along the coast, particularly in Mazandaran and Gilan provinces, Iran, also discharge waste directly into the Caspian Sea (4).

It is evident that seafood, particularly marine and freshwater fish, is a major component of the local diet. It is estimated that an average Iranian person, especially in Mazandaran province, consumes fish or shellfish three or more times per week.

The aim of this study was to survey organochlorines levels (D.D.T and its metabolite D.D.A) in four species of consumed fish in local markets in order to estimate potential human exposure.

MATERIALS AND METHODS

Eighty fish samples, Sefid (Rutilus frisikatum), Koli (Clupeonella delicatula), Kafal (Magila auratus) and Kilka (Vimba vimba), of approximately similar size, to minimize the effect of body weight, were collected manually using a cast net from 4 major fishing centers (Chalous and Babolsar cities, Khazar Abad and Miankaleh regions) in Mazandaran province, Iran in 2008. The samples were cleaned and stored in ice-cooled polyethylene bags for transportation to the laboratory.

The dorsal muscle tissues of the fish were homogenized separately in a high-speed blender until a paste-like consistency was formed (i.e. following the procedure of the US Food and Drug Administration) (9,16). After preparing the homogenate, 20
g of the sample was taken and mixed with 20 g of anhydrous sodium sulfate (Na$_2$SO$_4$). This mixture was, then, ground in a mortar until it reached the consistency of a free flowing powder. The powder was transferred to a paper thimble and extracted in a Sohulet apparatus using 250 ml of solvent (double distilled (DD) hexane: DD dichloromethane, 1:1) for 6 hours at 25°C. The extract was, then, evaporated to dryness to calculate the fat content. Then 0.3 g of fat was taken and cleaned up through glass column chromatography with deactivated florisil (mesh: 60-100 m) and the extract was eluted through a glass column with 100 ml eluting mixture (DD hexane: DD dichloromethane, 1:1). The extract was, then, evaporated to dryness and made to a final volume (5 ml) with DD hexane for evaluation of D.D.T and D.D.A levels using gas chromatography (GC) analysis.

**RESULTS**

The assessed concentrations of D.D.T and D.D.A residues in the four species of fish in the Caspian Sea are presented in Table 1. The results showed that mean concentration of *Rutilus frisikutum* fish had the highest level of D.D.T (0.029±0.003 mg/kg) in Babolsar sampling zone. In Chalus and Babolsar regions, *Clupeonella delicatula* fish had similar values of D.D.T (0.025±0.005 and 0.024±0.002 mg/kg, respectively). The samples of *Mugila auratus* fish in Chalus sampling center presented the maximum values of D.D.T (0.033±0.008 mg/kg) and finally the *Vimba vimba* fish presented the minimum value of D.D.T in comparison with the other species of fish collected from the sampling zones.

In addition, the D.D.A residues in the studied species of fish were found in the order of *Rutilus frisikutum* (0.031±0.007 mg/kg, Babolsar) > *Mugila auratus* (0.030±0.002 mg/kg, Chalus) > *Clupeonella delicatula* (0.026±0.002 mg/kg, Chalus) > *Vimba vimba* (0.019±0.001 mg/kg, Babolsar and Miankaleh) fish.

Statistical analysis (one-way ANOVA) indicated that there were no significant differences between D.D.T contents determined in the fish sampled from the sampling regions (P<0.05), but significant differences were observed in D.D.A values found in the fish samples (P<0.05).

**Table 1.** Concentrations of D.D.T and D.D.A in the fish sampled from the 4 sampling zones in the Caspian Sea in 2008

<table>
<thead>
<tr>
<th>Sampling zones</th>
<th>Fish species</th>
<th>D.D.T</th>
<th>D.D.A</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chalus</td>
<td><em>Rutilus frisikutum</em></td>
<td>0.024±0.002</td>
<td>0.026±0.001</td>
</tr>
<tr>
<td></td>
<td><em>Clupeonella delicatula</em></td>
<td>0.025±0.005</td>
<td>0.026±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Mugila auratus</em></td>
<td>0.033±0.008</td>
<td>0.030±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Vimba vimba</em></td>
<td>0.016±0.003</td>
<td>0.016±0.003</td>
</tr>
<tr>
<td>Babolsar</td>
<td><em>Rutilus frisikutum</em></td>
<td>0.029±0.003</td>
<td>0.031±0.007</td>
</tr>
<tr>
<td></td>
<td><em>Clupeonella delicatula</em></td>
<td>0.024±0.002</td>
<td>0.024±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Mugila auratus</em></td>
<td>0.022±0.002</td>
<td>0.022±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Vimba vimba</em></td>
<td>0.019±0.001</td>
<td>0.019±0.001</td>
</tr>
<tr>
<td>Khazar abad</td>
<td><em>Rutilus frisikutum</em></td>
<td>0.019±0.002</td>
<td>0.021±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Clupeonella delicatula</em></td>
<td>0.017±0.001</td>
<td>0.021±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Mugila auratus</em></td>
<td>0.022±0.002</td>
<td>0.015±0.001</td>
</tr>
<tr>
<td></td>
<td><em>Vimba vimba</em></td>
<td>0.016±0.001</td>
<td>0.017±0.001</td>
</tr>
<tr>
<td>MianKaleh</td>
<td><em>Rutilus frisikutum</em></td>
<td>0.018±0.001</td>
<td>0.019±0.001</td>
</tr>
<tr>
<td></td>
<td><em>Clupeonella delicatula</em></td>
<td>0.018±0.001</td>
<td>0.019±0.001</td>
</tr>
<tr>
<td></td>
<td><em>Mugila auratus</em></td>
<td>0.020±0.002</td>
<td>0.020±0.002</td>
</tr>
<tr>
<td></td>
<td><em>Vimba vimba</em></td>
<td>0.017±0.002</td>
<td>0.019±0.001</td>
</tr>
</tbody>
</table>

* The concentrations are based on Mean±SD (mg/kg); Number of fish samples (n=80)
DISCUSSION

The concentrations of D.D.T and D.D.A in the selected species of fish in each site were measured. Despite the importance and intensity of poisonous residues in food chain, there are little monitoring and few studies on fish in the Caspian Sea. Thus, conducting studies to ascertain the level of concentrations of current consumed pesticides in environment is of vital importance.

Due to lipo-solubility and bioaccumulation of OCs in the species of fish, as a member of human food chain, they should be monitored in the environment. Noticing the severe toxic effects, persistence of organic micro pollutants and trace elements, and concerning levels of contamination in aqua systems, especially in fish, officials are recommended to limit the consumption of pesticides which can contaminate the environment (16-18).

A study located in north Atlantic ocean indicated that concentration of DDT, DDE, Dieldrin, and Endosulphan in liver tissue of Shir bit fish samples were 0.002, 0.002, 0.006, and 0.007 mg/kg, respectively, that were lower than the standard quantities proposed by WHO (0.05 mg/kg) (19). Also, the measured quantities of D.D.T and D.D.A in the fish samples in the study were lower than WHO standard levels adjusted for safety of fish, but following the development of scientific knowledge, determination of more precise limits is necessary to improve public health. As a result, guidance of farmers to use pesticides or fertilizers, control of house wastewaters flowing into rivers and crop fields, and the establishment of reference laboratories should be performed (20).

CONCLUSION

The purpose of this study was to determine the concentrations of D.D.T and its metabolite D.D.A in four species of most consumed fish collected from the Caspian Sea.

The findings of this study showed that values of the heavy metals in the evaluated samples were within the standard limits set by various authorities. Finally, the following suggestions for safety improvement along with daily improvement of industries and industrial pollution are made (16,20):
- Frequent long-term measurement of pesticide residues in food products (agricultural and sea foods, etc)
- Establishment of reference laboratories equipped with analytical apparatuses, such as GC (gas chromatography), GC mass, HPLC (high performance liquid chromatography), etc.
- Informing people and farmers about the instructions for using pesticides, especially chlorinated pesticides, and controlling their probable toxicity;
- Evaluation of chlorinated pesticides in human organs and fluids (blood, milk, fat tissues, liver, etc.).

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