SciForschen
Open HUB for Scientific Research

Nutrition and Food Technology: Open Access

ISSN 2470-6086 | Open Access

RESEARCH ARTICLE

Volume 8 - Issue 2

Contaminations in Wild and Farmed Fish and Influence of Traditional Cooking Methods

Samar Aref[1,*], Noha Morsy[2], Ramadan A Habiba[1], Mohamed Abdel-Daim[3], and Fatma Zayet[1]

[1]Department of Food Science & Technology, Faculty of Agriculture, Suez Canal University, Ismailia, Egypt
[2]Department of Food Science & Technology (Home Economics), Faculty of Agriculture, Suez Canal University, Ismailia, Egypt
[3]Department of Pharmacology, Faculty of Veterinary Medicine, Suez Canal University, Ismailia, Egypt

***Corresponding author:** Samar Aref, Department of Food Science & Technology, Faculty of Agriculture, Suez Canal University, Ismailia, Egypt, Tel: 00201008522305; E-mail: Samar.moh.aref@gmail.com

Received: 22 Jun, 2022 | **Accepted:** 18 Jul, 2022 | **Published:** 25 Jul, 2022

Citation: Aref S, Morsy N, Habiba RA, Abdel-Daim M, Zayet F (2022) Influence of Traditional Cooking Methods on the Concentrations of some Contaminants in wild and farmed fish in Ismailia Governorate, Egypt. Nutr Food Technol Open Access 8(2): dx.doi.org/10.16966/2470-6086.181

Abstract

Due to the increasing concern of consumers about farmed fish contamination, in this study, different wild and farmed fish species (Nile tilapia (*Oreochromis niloticus*), gray mullet (*Mugil cephalus*), and thin lip gray mullet (*Liza ramada*) commonly consumed in Egypt, were chemically analyzed for the presence of heavy metals, antibiotics, polycyclic aromatic hydrocarbons, organochlorine pesticides residues, hormones and microbiologically evaluated for total counts of aerobic and coliform bacteria. Moreover, the influence of traditional cooking methods (i.e., grilling, frying, and baking) on these parameters were estimated. The results revealed that the polycyclic aromatic hydrocarbons and hormones were not detected in the fish species. However, heavy metals; cadmium, lead, nickel, cobalt, zinc, and mercury) were found in ranges of 0.014-0.35, 0.064-0.358, 0.027-0.875, 0.058-0.750, 0160-1.345 and 0.062-0.426 mg/kg, respectively. In addition, the organochlorine pesticides residues (p,p^1-DDE, p,p^1-DDT, and Chlorpyrifos) in the studied fish samples ranged between 0.02-2.13 mg/kg in the marine sites only. The thermal treatments *via* grilling were the most effective cooking method followed by baking to decrease the heavy metals concentrations in the fish species. Also, all detected pesticide residues and antibiotics in different fish species were decreased by cooking methods. Generally, the studied cooking methods significantly ($p<0.05$) decreased the total bacterial count and coliform counts in all treated samples by %. The frying method was the most effective cooking method in decreasing bacterial counts. In addition, neither *Escherichia coli* nor *Vibrio spp* was detected in the fish samples. Moreover, the studied cooking methods seemed to be suitable for decreasing the studied contaminants. Consequently, the farmed and wild fish species in the studied sites in Ismailia Governorate, Egypt, were safe and suitable for human consumption. Periodical monitoring of all sites for pollutants is highly recommended to ensure consumer protection.

Keywords: Contaminants; Farmed fish; Traditional cooking methods; Heavy metals; Bacterial counts

Introduction

Globally, aquaculture accounts for approximately half of the fish consumed and are considered among the quickest-growing food industries [1], with a growth rate of around 8.5% [2]. This trend is expected to continue, with aquaculture's contribution to fish food supply expected to reach 60% by 2020 [3]. Moreover, aquaculture is the world's quickest developing food production sector, with an annual ratio of growth of 37% registered in 2016 and an expected production of 109 million tons by 2030 [4].

Contaminants are usually present in natural environments as complex mixtures such as toxic heavy metals, Polycyclic Aromatic Hydrocarbons (PAHs), pesticides and bacterial contamination, providing many vital indicators for a complete diagnosis of environmental degradation. However, heavy metals are regarded as a severe threat to the aquatic environment because of their environmental stability and poisonous effects on aquatic organisms [5]. Heavy metal pollution can have destructive effects on the environment's biological balance and various aquatic creatures [6]. Moreover, it might be accumulated in many tissues and organs of fish species [7], which can enter into the human metabolism through consumption, causing serious health risks. Because of the nutritional value of fish, it is necessary to define the level of heavy metals in aquatic organisms and its suitability for human utilization [8,9].

Antibiotics used in treating animal diseases and promoting animal growth are extensively used in fish feeds, and their presence is rising. For instance, sulfonamides and tetracycline are two classes of antibiotics generally utilized in aquaculture to treat infections in fish [10]. The excessive use of anti-microbials in aquaculture leads to antibiotic residue in consumed fish and fish products. This results in the undetected intake of antibiotics by the consumers with the probable changing of their normal flora which raises their vulnerability to infectious diseases by bacteria and to choose for

antibiotic-resistant microbes [11]. Undetected antibiotic intake in food can be poisonous and cause allergy [12,13]. Therefore, there is a worldwide concern around the consumption of seafood having even low rates of antibiotics.

Similarly, PAHs are common organic contaminants in the environment that are recognized for their carcinogenic and mutagenic impacts and their ability to bioaccumulate in animal and human tissue [14]. PAHs are categorized as environmentally hazardous contaminants because of their known mutagenic, lipophilic and carcinogenic properties [15], in addition to their endocrine disrupting action [16]. In fish, PAHs have been known to be poisonous after biotransformation *via* poisonous metabolites that could be tied covalently to cellular macromolecules like RNA, DNA, and proteins, resulting in carcinogenesis, mutagenesis and cell damage [16]. PAHs reach the marine environment mainly by atmospheric depositions and surface runoff, where they may pollute aquaculture sediments and subsequently enter the food chain *via* fish to people [17]. In addition, Anderson, et al. [18] have reported that dietary exposure to PAHs is related to some human cancers.

Organochlorine Pesticides Residues (OCPs) are synthetic organic components with strong bonds among their chlorine and carbon compounds. Organochlorine pesticides are used for controlling pests, undesirable species of animals or plants causing harm or interfering with the production, storage, processing, and marketing of food, and agricultural products. It is important to remember that all pesticides must be regarded as active toxins [19]. OCPs are hydrophobic and steady components. Their photo-oxidation, low vapor pressure, and low chemical decomposition levels, have led to their accumulation in biological tissues and consequently enlargement in living organisms *via* food chains. Muir D, et al. [20] reported that biological samples like fish, terrestrial mammals, and sediments have greater OCPs than air or water making them more appropriate for routine monitoring of organochlorine residues and more important in the context of people's exposure. The adverse effects of pesticide residues in fish include acute and persistent damage to the nervous system, damage to the reproductive organs, and dysfunction of the immune systems [21]. Many of persistent organochlorines and extremely poisonous organophosphates, that have been prohibited or extremely limited, are still marketed and used in many developing countries. Misuse of pesticides by individuals and weak national controlling plans are behind the negative impacts of these chemicals in developing countries [22].

Hormones are used for sex reversal and in artificial reproduction. The first is used when the growth rate is different among males and females. The second retains the production chain with the continuous production of eggs. This contrast among gender is commonly present in teleost fish and often happens during sexual maturity [23]. However, the usage of hormones in fish cultivating can have negative impacts, like possible environmental hazards and human health associated with hormone-dependent factors. Besides, its utilization exterior of good animal cultivation practices can impact the fish farming production system and the commercialization of this food product. The synthetic steroid 17 α-methyltestosterone is a particular male hormone commonly used to initiate sex reversal in fish [24]. The addition of steroids to the water containing sexually undifferentiated fish has also been successful in changing sex ratios and can provide aquaculturists with a safe and cost-effective to treat fry with food that contains 17α-methyltestosterone. The impact of these residues is great on human health since it may cause adulthood for girls and boys, carcinoma, increased embryo mortality, and liver tumors [25]. Sex steroid hormones can cause cancer in particular target organs (mammary gland, uterus, cervix, and prostate gland) and are characterized as carcinogens [26].

The pollution of fish with pathogenic bacteria from the aquatic environment may be a risk in all fisheries [27]. The level of fish pollution at the time of capture will depend on the microbiological quality of the water in which fish are caught. Many parameters will affect the bacteria such as water temperature, salt content, the proximity of harvesting regions to human habitations, amount, and source of fish food, and method of harvesting. The consumable flesh of the fish is ordinarily sterilized at the moment of capture, but bacteria are often growing on the gills, skin, and intestinal tract. In addition, fish take many microbes into their digestive tract from water, sediment, and food [28].

Generally, risk estimates for chemical residues depend on residue levels in raw food, even though a major amount of food is cooked or processed before consumption. To effectively evaluate the hazards of contaminants residues to humans, the impacts of cooking procedures (grilling, baking and frying) must be investigated [29]. Cooking methods are known to reduce the hazard of contaminants in fish. The decrease in the heavy metal concentrations is influenced by cooking procedures because of the discharge of these metals with the loss of drip, probably in relation to dissolvable amino acids and un-coagulated proteins bounded with metals. The cooking process may reduce the protein content of the fish parts [30]. Also, lipophilic OCPs are associated with the fat portion of foods. Hence, cooking methods that release or eliminate fat from fish will tend to decrease the total amount of OCPs in the cooked fish [31]. Furthermore [32] stated that heat treatment may diminish the antimicrobial activity of the antibiotic residues.

Therefore, the main objective of the study was to determine the level of some contaminants such as heavy metals, polycyclic aromatic hydrocarbons, organochlorine pesticides residues, hormones, antibiotics as well as total counts of aerobic and coliform bacteria. Moreover, the influence of traditional cooking methods (i.e., grilling, frying, and baking) on some contaminants in the flesh of wild and farmed fish species; commonly consumed in Ismailia Governorate, Egypt; to remove the consumer suspicious about the safety of fish consumption obtained from the studied sites.

Materials and Methods

Study areas and samples collection

Farmed Nile tilapia (*Oreochromis niloticus*) was collected from three fish farms in different sites of Ismailia Governorate: Farm 1 (F1), a private fish farm at Altal AlKabeir city, Farm 2 (F2) is a private fish farm at El Qantara Gharb region, Ismailia Governorate, and Farm 3 (F3), Institute of Aquaculture and Fish Technology of Suez Canal University. Moreover, wild Nile tilapia (*Oreochromis niloticus*) was also collected from freshwater (*Ismailia Canal*) (FW) and saltwater (*Lake Timsah*) (MW1). Also, the flathead gray mullet (*Mugil cephalus*) (MW2) and the thin lip gray mullet (*Liza ramada*) (MW3) were collected from Lake Timsah.

Fish samples having approximately similar size (average bodyweight 150-200g) were collected during the study period. The time and date of harvest were the same for both farmed and wild fish in each location but varied among locations. Immediately after collection, fish were kept in an icebox with a sufficient amount of flake ice and transferred to the laboratory. After collection, samples were transported to the Central Laboratory of Residue Analysis of Pesticides and Heavy Metals in Food (QCAP Lab) and kept frozen at −20°C until the time of

carrying out the physicochemical and microbial assays.

Traditional cooking methods

Whole fish samples obtained from each farm, freshwater, and saltwater were divided into three groups then cooked by grilling, frying and baking methods using the methods described by El-Sherif SA, et al. [33]. For grilling, fish samples were pre-coated first with bran (a local routine) and grilled using an electrical grill machine at 180°C for 10 min. In the frying method, fish samples were pre-coated first with wheat flour (a local routine) and then fried at 160°C in deep sunflower oil in a frying pan for 8 min. Fish samples were placed on a metal tray and baked in an electric oven at 180°C for 20 min.

Analytical methods

Chemical analysis: Heavy metals involving Nickel (Ni), Cadmium (Cd), Cobalt (Co), Lead (Pb), Zinc (Zn), and Mercury (Hg) were determined *via* an Atomic Absorption spectrophotometer (Thermo Electron Corp, S series, AA spectrometer, Type S4 AA system, China) according to AOAC [34]. Total mercury was measured by an inductively coupled plasma mass spectrometry (ICP-MS) according to Bloxham MJ, et al. [35]. Antibiotics were determined by LC-MS according to the method described by Shin D, et al. [36]. PAHs were determined by GC according to Almatari MS, et al. [37]. Organochlorine Pesticides (OP's) residues were determined using GC-MS according to the modified QuEChERS sample preparation method described by Molina-Ruiz JM, et al. [38]. Hormones were measured by GC-MS according to Seo J, et al. [39].

Microbiological analysis: Ten grams from the blended sample were aseptically taken out of the Petri dish where 90 mL sterile buffered peptone water was subsequently added. Samples were then homogenized for 2 min. Aerobic Plate Count (APC) was determined using the pour plate method. Serial dilutions were prepared, and then, 1 mL of each was placed onto plate count agar media (Merck, Darmstadt, Germany). Then, the plates were incubated at 35°C for two days. The number of *Vibrio species*, 1.0 mL from each dilution was plated on Thiosulfate Citrate Bile Salts Sucrose (Oxoid) agar plates by the pour plate method. The inoculated plates were incubated for 18- 24 h at 37°C. For total coliform count, violet red bile agar was used as a medium (approximate formula per liter: 3g yeast extract, 7 g Peptone, 1.5 g bile salts, 10 g Lactose, 5 g sodium chloride, 0.03 g neutral red, 2.0 mg crystal violet and 15 g agar) plates were incubated at 35°C for 18-24 h. After incubation, the typical purple round bacterial colonies were enumerated. For Fecal coliform and *E. coli*, typical purple colonies were counted and were further confirmed by growing in Eosin Methylene Blue (EMB) agar plates. Presumptive colonies (blue-black colonies with a green metallic sheen and dark centers) on EMB agar were streaked on slant agar. Results are expressed as log CFU per gram of sample.

Statistical analysis: Analysis of variance was done using SPSS software (version 16.0 for Windows, SPSS Inc., Chicago). Bartlett's multiple range tests were used to locate significance between treatment means at $P \leq 0.05$.

Results and Discussion

Chemical analysis

Heavy metals analysis: The concentrations of heavy metals (Cd, Pb, Ni, Co, Zn and Hg) (mg kg-1) of weighted tissues of wild and farmed fish collected from the Ismailia Governorate are presented in figure 1. The highest Cadmium (Cd) concentration in the muscle of (0.083 mg/kg) was observed in marine Mullet (*Liza ramada*), while the lowest level (0.014 mg/kg) was detected in freshwater Nile tilapia. The data revealed a significant difference ($P \leq 0.05$.) in Cd values between different fish samples at Ismailia Governorate. The values of Cd concentrations observed in fish samples agree with those values reported in the most reviewed literature, which reports Cd values in fish from the Pearl River Delta ranged from 0.03 to 1.57 mg/kg [40], from 0.01 to 0.04 mg/kg in fish from Fujian Province [41], and from 0.024 to 0.030 mg/kg in fish from China's Yellow River [42]. To be specific [43] detected Cd mean value of 0.034 mg/kg in muscle tissues of tilapia, which is similar to the results reported in this study. According to the results recorded in the figure 1, Cd values were within the permissible limit (0.1 mg/kg in muscle) stipulated by WHO/FAO.

The mean concentrations (mg/kg) of Lead (Pb) in muscles of various fish species in this study ranged from 0.064 to 0.358 mg/kg. Marine Nile tilapia (*Oreochromis niloticus*) of the Institute of Aquaculture and Fish Technology (F3) had the highest Pb concentration (0.358 mg/kg), whereas the marine Mullet (*Liza ramada*) showed the lowest concentration (0.064 mg/kg). By the way, Pb was not detected in El Qantara Gharb farm (F2) and freshwater Nile tilapia (FW1) samples in this study (Figure 1). In general, Pb concentrations measured in all fish samples collected from different locations were below the permissible limit (0.3 mg/kg in muscle) stipulated by WHO/FAO except for fish samples collected from F3.

The lowest concentration of Nickel (Ni) in fish species was 0.027 (mg/kg) found in Nile tilapia from Altal AlKabeir farm, while the highest one of 0.875 (mg/kg) was observed in marine Mullet (*Mugil cephalus*). The sequence of concentration levels from the highest to the lowest for Ni was detected as follows: MW1>MW3>FW>F3>MW2>F1. Fortunately, Ni was not detected in fish samples from Qantara Gharb farm (F2). The data revealed a significant difference ($P \leq 0.05$.) in Ni values between wild and farm fish samples at Ismailia. In addition, the concentration of Ni in fish samples collected in this study was within the range of Ni concentration (0.02 to 3.97 mg/kg) in a similar study reported by Türkmen M, et al. [44] in fish from the Marmara, Aegean, and Mediterranean seas. However, the results of Ni values, presented in figure 1, exceed the permissible limit (0.02 mg/kg in muscle) stipulated by WHO/FAO.

According to the results in figure 1, the range of concentration of Cobalt (Co) in fish muscles was 0.028-0.750 mg/kg in all studied fish species. The level of Co concentration was significantly higher ($P \leq 0.05$.) in wild fish samples compared to farmed fish samples. The sequence of concentration levels from the highest to the lowest for Ni detected was as follows: MW3>MW2>MW1>FW>F1>F2>F3. In general, Co element concentration in all fish samples investigated in this study from different sources values did not exceed the permissible limit (1 mg/kg in muscle) stipulated by WHO/FAO.

The data in figure 1 showed that Zinc (Zn) was detected in all wild and farmed fish samples examined in this study. The level of Zn concentration in fish muscle ranged from 0.160 mg/kg for Nile tilapia from Altal AlKabeir farm to 1.345 mg/kg for marine Mullet (*Mugil cephalus*). The data revealed that there were significant differences ($P \leq 0.05$.) in Zn values between wild and farmed fish samples at Ismailia Governorate, and all of them were higher than the permissible limit (0.04 mg kg^{-1}) stipulated by WHO/FAO.

Finally, Mercury (Hg) was detected only in marine species ranging from 0.062 mg kg-1 to 0.426 mg kg-1. Fortunately, the levels of Hg concentration in all fish species were lower than the permissible limits of WHO/FAO. Comparable, results were reported by Morshdy A, et al. [45] were observed in Brush tooth Lizardfish and Mackerel fish

samples collected from Egypt with Hg concentrations from 0.54 ± 0.03 to 0.56 ± 0.01 ppm, respectively.

Antibiotics and PAHs: In this study, out of 30 monitored antibiotics in fish samples, only two (Chlortotetracycline and Doxycycline) were detected in farmed fish. Doxycycline values ranged from undetected in F2 farm to 26 µg/kg in F1 and reached up to 35 µg/kg in F3. Tetracycline residues were only found in flesh fish of F3 farm (Table 1). Choi J, et al. [46] studied antibiotic residues in seven farmed fish species in Korea, and they detected only enrofloxacin (0.0305 mg/kg) in *Cyprinus carpio*. Done HY, et al. [47] investigated 47 types of antibiotics in diverse cultivated fish (rainbow trout, Atlantic salmon, tilapia, and catfish) and shrimp retailed in the US. They identified five antibiotics (virginiamycin, sulfadimethoxine, epioxytetracycline, oxytetracycline, and ormetoprim) exceeding the detection limits. Barani A, et al. [48] found a high occurrence of tetracycline (63.1%), enrofloxacin (19.6%), florfenicol (40.6%), and sulfamethazine (17.4%) in rainbow trout samples from Iranian trout farms. On the other hand, PAHs as a common organic contaminant in the environment were not detected in any fish sample in this study.

Pesticide residues and Hormones: Data collected from this study revealed that only three organochlorine pesticides OCPs were detected in fish samples from marine and freshwater (p,p′-DDE, p,p′-DDT, and Chlorpyrifos). The data indicated that, p,p^1-DDE, and Chlorpyrifos range values were 0.02-2.01, undetected-2.044, and 2.05 to 2.13 mg/kg, respectively in Nile tilapia and marine Mullet in both salt and freshwater (Table 2). Also, the levels of OCPs in freshwater fish samples were lower than that in marine fish samples. The marine fish coming from the industrialized regions might have larger pollution rates. Previous research on wild tilapia has showed that DDE is the most prominent RDDT metabolite in fish tissue [49]. Yamashita N, et al. [50] reported that DDE was the most common pesticide residue in fish samples obtained from the Nile and Manzala lakes (7.6-67 ng/g).

Fortunately, hormones were not detected in any wild or farmed fish samples in this study. It should be noted that, fish and fish products that contribute to in human nutrition should be secure and free of potentially dangerous ingredients which affect human health [51].

Microbiological analysis

Fish is among the most perishable foods, primarily because of the activity of microorganisms on the surface of the freshly caught fish. Bacterial growth is commonly considered the primary reason for fish and fish product deterioration. As a result, it is suggested that bacterial count could be used as a quality indicator for fish products [52].

Table 1: Antibiotics concentrations (µg/kg) detected in farmed fish muscle.

	Antibiotics	
	Chlortetracycline	Doxycycline
F1	<LOQ	26^b
F2	ND	ND
F3	36	35^a

F1: Altal AlKabeir farm, F2: El Qantara Gharb farm, and F3: Institute of Aquaculture and Fish Technology.

ND=Not detected or the metals exist in amounts below the limit of detection.

LOQ: Limit of Quantification.

*Means within the same column having different superscripts are significantly different at P ≤ 0.05.

Aerobic Plate Counts (APC) are used as an acceptability index in fish products. As shown in table 3, it could be noticed that the low number of APC in the examined fish species demonstrated great safety and remained within the permitted limits which did not exceed 10^6 cell/g as stated by EOS [53] following Shen L, et al. [54], who documented that the total count of fresh fish <10^4 cells/g, sub fresh 104-106 and deteriorated fish >10^6 cells/g sample. The data revealed no significant differences (P ≤0.05.) in APC values between different fish samples. The values of bacterial counts were lower than those reported by Mohamed FSA, et al. [55], which showed that the total bacterial count in raw Nile tilapia was 2.11 × 10^5 cfu/g and 2.02 × 10^5 cfu/g sample.

The average count of coliform bacteria of the examined fish samples ranged from 3.49 to 5.48 log cfu/g. Because coliforms are not part of the natural bacterial flora in fish, their appearance shows the contamination rate of their ecosystem. In addition, the data showed no significant difference in coliform values between different fish samples.

Table 2: Pesticide residues concentrations (mg/kg) in farmed and wild fish muscle.

	p, p^1-DDE	p, p^1-DDE	Chlorpyrifos
F1	ND	ND	ND
F2	ND	ND	ND
F3	ND	ND	ND
FW	<LOQ	ND	0.05^b
MW1	0.02^c	<LOQ	<LOQ
MW2	1.01^b	1.067^b	<LOQ
MW3	2.01^a	2.044^a	2.13^a

F1: Altal AlKabeir farm, F2: El Qantara Gharb farm, F3: Institute of Aquaculture and Fish Technology, FW: freshwater fish, MW1: marine Nile tilapia (Oreochromis niloticus), MW2: marine Mullet (Mugil cephalus), and MW3: marine Mullet (Liza ramada).

ND=Not detected or the metals exist in amounts below the limit of detection.

LOQ: Limit of Quantification.

*Means within the same column having different superscripts are significantly different at ≤0.05.

Table 3: Bacterial count (log cfu/g) in farmed and wild fish muscle.

	APC	Coliform	*E. coli*	Vibrio
F1	5.17^a	3.49^a	ND	ND
F2	5.65^a	5.31^a	ND	ND
F3	5.20^a	4.96^a	ND	ND
FW	5.58^a	5.48^a	ND	ND
MW1	5.25^a	4.34^a	ND	ND
MW2	5.79^a	4.50^a	ND	ND
MW3	5.56^a	4.52^a	ND	ND

F1 Altal AlKabeir farm, F2 El Qantara Gharb farm, F3 Institute of Aquaculture and Fish Technology, FW1 freshwater fish, MW1 marine Nile tilapia (*Oreochromis niloticus*), MW2 marine Mullet (*Mugil cephalus*), and MW3 marine Mullet (*Liza ramada*).

Means within the same column having different superscripts are significantly different at P ≤ 0.05.

Table 4: Effect of traditional cooking methods on heavy metals concentrations (mg/kg) detected in farmed and wild fish muscle.

Element	Treatment	F1	F2	F3	FW	MW1	MW2	MW3	Mean
Cd	Raw	0.015^{bc}	ND	<LOQ	0.014^{bc}	0.034^{b}	0.029^{b}	0.083^{a}	
	Grilling	ND	ND	ND	ND	ND	ND	<LOQ	0^{A}
	Frying	0.21^{a}	ND	ND	ND	ND	ND	<LOQ	0.03^{A}
	Baking	ND	ND	ND	ND	ND	ND	<LOQ	0^{A}
Mean		0.07^{A}	0^{A}	0^{A}	0^{A}	0^{A}	0^{A}	0^{A}	
Pb	Raw	0.169^{ab}	ND	0.358^{a}	ND	0.086^{ab}	0.097^{ab}	0.064^{ab}	
	Grilling	<LOQ	ND	<LOQ	ND	ND	0.723^{b}	ND	0.10^{B}
	Frying	<LOQ	ND	0.432^{c}	ND	<LOQ	1.234^{a}	ND	0.23^{A}
	Baking	<LOQ	ND	<LOQ	ND	<LOQ	0.834^{b}	ND	0.11^{B}
Mean		0^{C}	0C	0.14^{B}	0^{C}	0^{C}	0.93^{A}	0^{C}	
Ni	Raw	0.027^{c}	ND	0.478^{b}	0.567^{b}	0.875^{a}	0.467^{b}	0.654^{b}	
	Grilling	<LOQ	ND	0.140^{b}	0.436^{b}	0.653^{b}	0.345^{b}	0.356^{b}	0.27^{A}
	Frying	<LOQ	ND	0.189^{b}	0.421^{b}	0.678^{b}	0.347^{b}	0.365^{b}	0.32^{A}
	Baking	<LOQ	ND	0.245^{b}	0.510^{b}	0.743^{a}	0.365^{b}	0.371^{b}	0.31^{A}
Mean		0^{A}	0^{A}	0.19^{A}	0.45^{A}	0.67^{A}	0.76^{A}	0.36^{A}	
Co	Raw	0.074^{c}	0.058^{c}	0.028^{c}	0.234^{b}	0.650^{a}	0.743^{a}	0.750^{a}	
	Grilling	<LOQ	<LOQ	ND	<LOQ	0.387^{c}	0.293^{c}	0.374^{c}	0.15^{A}
	Frying	<LOQ	<LOQ	ND	<LOQ	0.431^{bc}	0.298^{c}	0.832^{a}	0.22^{A}
	Baking	<LOQ	<LOQ	ND	<LOQ	0.452^{bc}	0.264^{c}	0.654^{ab}	0.19^{A}
Mean		0^{C}	0^{C}	0^{C}	0^{C}	0.42^{B}	0.28^{B}	0.62^{A}	
Zn	Raw	0.160^{c}	0.563^{bc}	0.214^{c}	0.865^{ab}	0.672^{bc}	0.942^{ab}	1.345^{a}	
	Grilling	0.033^{c}	0.378^{bc}	0.021^{c}	0.476^{bc}	0.382^{bc}	0.487^{bc}	0.832^{bc}	0.37^{A}
	Frying	0.101^{bc}	0.389^{bc}	0.301^{bc}	0.432^{bc}	0.465^{bc}	0.567^{bc}	1.905a	0.59^{A}
	Baking	0.185^{bc}	0.412^{bc}	0.156^{bc}	0.561^{bc}	0.498^{bc}	0.593^{bc}	1.04^{ab}	0.49^{A}
Mean		0.10^{B}	0.39^{B}	0.15^{B}	0.48^{B}	0.44^{B}	0.54^{B}	0.25^{A}	
Hg	Raw	ND	ND	<LOQ	ND	0.062	0.353^{a}	0.426^{a}	
	Grilling	ND	ND	ND	ND	ND	<LOQ	ND	0^{C}
	Frying	ND	ND	ND	ND	ND	<LOQ	ND	0^{C}
	Baking	ND	ND	ND	ND	ND	<LOQ	ND	0^{C}
Mean		0^{C}	0^{C}	0^{C}	0^{C}	0^{C}	0^{C}	0^{C}	

F1: AlTal AlKabeir farm, F2: El Qantara Gharb farm, F3: Institute of Aquaculture and Fish Technology, FW1: freshwater fish, MW1: marine Nile tilapia (*Oreochromis niloticus*), MW2: marine Mullet (*Mugil cephalus*), and MW3: marine Mullet (*Liza ramada*).

ND=Not detected, or the metals exist in amounts below the limit of detection.

LOQ: Limit of Quantification.

Means within the same column having different superscripts are significantly different at $P \leq 0.05$ for treatments, while those within the same row with different superscripts are significantly different for farms.

Results in table 1 revealed that neither *E. coli* nor *vibiro spp* was detected in the investigated wild and farmed fish samples, indicating that the wild and farmed fish (Nile tilapia and Mullet) were obtained from Ismailia Governorate were microbiologically safe and had better quality for human consumption.

The effect of traditional cooking methods on some contaminants

The effect of traditional cooking methods on heavy metals: The effect of traditional cooking methods on heavy metals (Cd, Pb, Ni, Co, Zn, and Hg) (mg kg^{-1}) detected in farmed and wild fish muscle are shown in table 4. Cadmium concentration in farmed tilapia was 0.015 mg/kg and increased to 0.21 ppm by frying, while Cd was undetected when samples were either grilled or baked. These results of Cd concentrations in this study were in agreement with those found by El-Sherif SA, et al. [33], who observed that Cd levels in uncooked Nile tilapia (0.058 ppm) did not change when fish samples were cooked by frying (0.058 ppm), and reduced to 0.034 ppm after grilling.

The lead concentrations were reduced from 0.097 of marine Mullet (*Mugil cephalus*) flesh to 0.834 (mg/kg) by baking and 0.723 by grilling. However, frying increased Pb concentration to 1.234 and

Table 5: Effect of traditional cooking methods on bacterial count (log cfu/g) in farmed and wild fish muscle.

Bacterial Count	Treatment	F1	F2	F3	FW	MW1	MW2	MW3	Mean
APC	Raw	5.17^{a}	5.65^{a}	5.20^{a}	5.58^{a}	5.25^{a}	5.79^{a}	5.56^{a}	
	grilling	4.34^{a}	4.80^{a}	4.51^{a}	4.76^{a}	4.47^{a}	3.93^{a}	5.10^{a}	4.55^{A}
	Frying	3.86^{a}	4.78^{a}	4.26^{a}	4.47^{a}	4.21^{a}	3.11^{a}	4.48^{a}	4.16^{A}
	Baking	4.51^{a}	5.35^{a}	4.67^{a}	5.34^{a}	4.67^{a}	4.12^{a}	5.34^{a}	4.85^{A}
Mean		4.23^{a}	4.97^{a}	4.48^{a}	4.84^{a}	4.45^{a}	3.72^{a}	4.97^{a}	
Coliform	Raw	3.49^{a}	5.31^{a}	4.96^{a}	5.48^{a}	4.34^{a}	4.50^{a}	4.52^{a}	
	Grilling	3.24^{ab}	4.84^{a}	4.51^{ab}	4.78^{a}	3.38^{ab}	4.45^{ab}	4.52^{ab}	4.25^{A}
	Frying	2.74^{b}	4.64^{ab}	4.21^{ab}	4.58^{ab}	3.16^{ab}	3.72^{ab}	4.52^{ab}	3.93^{A}
	Baking	3.27^{ab}	5.10^{a}	4.72^{a}	4.89^{a}	3.54^{ab}	4.39^{ab}	4.52^{ab}	4.34^{A}
Mean		3.10^{C}	4.86^{A}	4.48^{AB}	4.75^{A}	3.36^{BC}	4.18^{ABC}	4.52^{A}	

F1: AlTal AlKabeir farm, F2: El Qantara Gharb farm, F3: Institute of Aquaculture and Fish Technology, FW1: freshwater fish, MW1: marine Nile tilapia (*Oreochromis niloticus*), MW2: marine Mullet (*Mugil cephalus*), and MW3 marine Mullet (*Liza ramada*).

ND=Not detected, or the metals exist in amounts below the limit of detection.

LOQ: Limit of Quantification.

Means within the same column having different superscripts are significantly different at P ≤0.05 for treatments, while those within the same row with different superscripts are significantly different for farms.

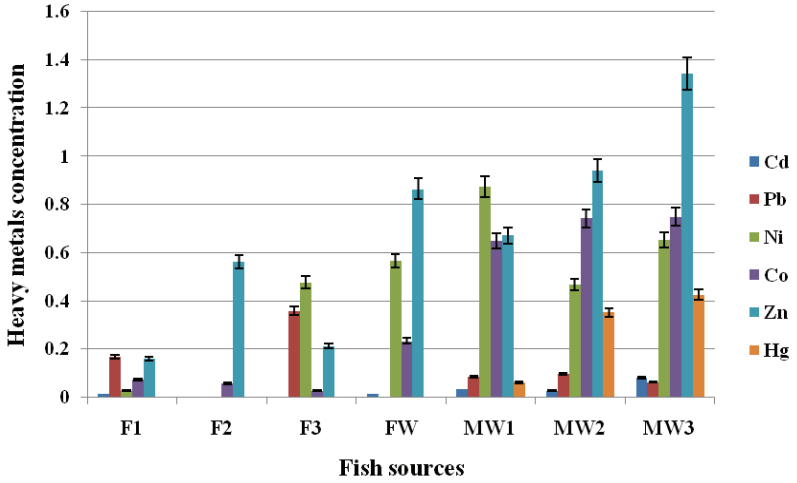

Figure 1: Heavy metals (mg/kg) concentrations detected in farmed and wild fish muscle.

F1: AlTal AlKabeir farm, F2: El Qantara Gharb farm, F3: institute of aquaculture and fish technology, FW1: freshwater fish, MW1: marine Nile tilapia (*Oreochromis niloticus*), MW2: marine Mullet (*Mugil cephalus*), and MW3: marine Mullet (*Liza ramada*). ND=Not detected, or the metals exist in amounts below the limit of detection.

LOQ: Limit of Quantification.

Means within the same column having different superscripts are significantly different at P>0.05 for treatments, while those within the same row with different superscripts are significantly different for farms.

0.432 (mg/kg) for Mullet (*Mugil cephalus*) and Nile tilapia. These findings agreed with those results reported by El-Sherif SA, et al. [33] in grilling Nile tilapia and mullet fish fillets from Wadi El-Rayan Lake, but differentiated in frying, where Pb concentrations were 0.415 and 0.196 ppm for uncooked Nile tilapia and mullet fish fillets, and reduced to 0.373 and 0.145 in grilled and 0.388 and 0.185 in fried samples, respectively. Also, the results were in agreement with those stated by Perello G, et al. [56], who revealed that the concentrations of Pb in tuna and sardine were higher in fried samples and lower in grilled samples. Therefore, the decrease in trace minerals levels as influenced by cooking procedures might be attributed to the release of these coagulated proteins bounded with metals, whereas the rise in minerals is associated with a reduction in the moisture content during cooking [57].

Also, Ni and Co concentration of examined fish samples decreased significantly by grilling followed by baking. Conversely, frying increased Ni and Co values in some fish samples.

It was noticed that Zn concentrations of marine Nile tilapia were 0.672 mg/kg, decreased to 0.465, 0.498, and 0.382 (mg/kg) for fried, baked, and grilled samples, respectively. El-Sherif SA, et al. [33] reported that Zn levels were 0.221 and 0.086 ppm in Nile tilapia and

mullet fish fillet from Wadi El- Rayan Lake rose to 0.263 and 0.116 ppm by frying method and reduced to 0.089 and 0.038 ppm by grilling method.

Total Mercury (Hg) concentrations found in the muscle of the studied fish samples revealed that Hg concentrations were below the maximum allowed for fish both in raw and cooked fish (0.5 mg kg). This implies that raw and cooked fish meet the existing safety criteria for wild and farmed fish at Ismailia Governorate. Furthermore, these findings suggested that, depending on the fish species, certain culinary procedures could help reduce total mercury levels in fish.

Therefore, it could be concluded that the concentrations of all determined heavy metals in the examined samples were lower than the permissible limit set by FAO/WHO [58]. Furthermore, grilling was the most effective method for decreasing all heavy metals, followed by baking. In contrast, frying method increased the majority of them. Therefore, cooking by grilling followed by baking methods was found to be more suitable for human consumption than the frying method for decreasing the harmful effects of heavy metals.

As there were only two antibiotics (Chlortetracycline and Doxycycline) detected in farmed fish, the antibiotics concentrations of baked, grilled, and fried fish samples were not detectable (below limits of detection) Uno K, et al., [59] reported that OTC residues were lower in all cooked samples than in the corresponding raw samples. Also [60] stated that application of heat treatment like roasting and frying may lead to destroying of all drug residues.

The effect of traditional cooking methods on pesticide residues: Because only three organochlorine pesticides were detected in the fresh fish samples from marine and freshwater (p,p′-DDE, p,p′-DDT, and Chlorpyrifos). Therefore, the concentrations of OCPs residues were investigated in cooked (grilled, fried, and baked) samples. OCPs residues concentration were not detected (data were not shown) after heat treatments of samples (Reduction rate 100% in most treatments). This result is supported by the results of Amal ME, et al. [61], who documented that OCPs levels (α-HCH, γ-HCH, and Endrin) in *Tilapia nilotica* and *Clarias lazera* fish taken from fish markets in Gharbia, Egypt were reduced after frying, grilling, and boiling. In *Tilapia nilotica* samples, OCPs concentrations were reduced by about 85, 79, and 80% by frying method, and 87, 52, and 100% by grilling, respectively. In *Clarias lazera* samples, the OCPs concentrations were reduced by 100, 54, and 83% by frying and 100, 50, and 100, respectively, by boiling.

Comparable results were detailed by Hassanen FS, et al. [62], who studied the effect of the grilling cooking method on OCPs concentrations in raw Nile tilapia (*Oreochromus nilotica*) collected from three different areas. The results showed a significant decrease in OCPs concentrations by a ratio of 11% to 100% by grilling depending upon the type of pesticide residue and variation in localities.

Domingo JL [63] explained that the reduction of OCPs residues by frying was because of increased loss of fats and oils with high heat treatment, which caused OCPs to be drained out with the lost fats and oils and in oil dripping by grilling. Furthermore, the decrease rate in OCPs residues in cooked fish varied relying on pesticide solubility, fish species, and cooking procedure [31].

The effect of traditional cooking methods on bacterial counts: The effect of traditional cooking methods on bacterial count (log cfu/g) in fish samples is shown in table 5. APC and coliform counts were reduced in all cooked fish samples. APC of raw *Mugil cephalus* was 5.79 and decreased to 3.93, 3.11, and 4.12 (log_{10} cfu/g) when grilling, frying, and baking were used, respectively. The APC of marine Nile tilapia (*Oreochromis niloticus*) flesh was 5.25 and decreased to 4.47, 4.21, and 4.67 when grilling, frying, and baking were used, respectively. This reduction in bacterial counts might be attributed to heat treatment applied during cooking methods. So, the greatest reduction was noted in fried samples compared with raw fish samples, followed by grilled and baked fish samples. These findings agreed with those published by El-Sherif SA, et al. [33] who found that APC of raw tilapia and mullet fish were 2.35 and 2.01 respectively, reduced to 1.95, 2.10, and 2.15 for tilapia fish and to 1.70, 1.72, and 1.95 (log_{10} cfu/g) for mullet by frying, grilling, and boiling, respectively.

Conclusion

In This study, the obtained results showed that most determined heavy metals, antibiotics, PAHs, OCPs, concentrations, and microbiological counts in flesh of wild and farmed fish species obtained from Ismailia Governorate were lower than the permissible limits. Hormones were not detected. Cooking methods further reduced contaminants; grilling being the most method for reducing heavy metals, antibiotics, and OCPs, followed by frying and baking. Furthermore, frying was the most effective method for reducing APC, followed by grilling and baking. Therefore, the traditional cooking methods (grilling, frying and baking) were suitable for decreasing the studied hazardous pollutants. Hence, the wild and farmed fish species obtained from the studied sites in Ismailia, Egypt were safe for human consumption removing the consumer suspicious about pollution of farmed fish as compared to wild fish in the studied area. Periodical monitoring of all sites for pollutants is highly recommended to ensure consumer protection.

Data Availability

All the authors have confirmed that the data supporting the results obtained from this study are present in this article.

Declarations

Conflict of interest: The authors declare no competing interests.

References

1. Subasinghe R, Soto D, Jia J (2009) Global aquaculture and its role in sustainable development. Rev Aquac 1: 2-9.
2. FAO (2013) FAO Fisheries and Aquaculture Department has published the Global Aquaculture Production Statistics for the year.
3. Washington S, Ababouch L (2011) Private standards and certification in fisheries and aquaculture: Current practice and emerging issues. FAO Fisheries and Aquaculture Technical Paper, No. 553: 181.
4. FAO (2018) National Aquaculture Sector Overview, Egypt. Fisheries and Aquaculture Department, Food and Agriculture organization of the United Nations, Rome, Italy.
5. Radwan AM, Khalil MK, El-Moselhy KM (2007) Distribution of phosphorus fractions and some of heavy metals in surface sediments of burullus lagoon and adjacent mediterranean sea. Egypt J Aquat Res 33: 277-289.
6. Vosylienė M, Jankaitė A (2006) Effect of heavy metal model mixture on rainbow trout biological parameters. Environmental Science 4: 12-17.
7. Heba HM (2004) Distribution of trace elements in the white muscles and some key organs of fish species from Al-Hodiedah Red Sea Coast of Yemen. Proc I C BS 3: 435-454.
8. Bosch AC, O'Neill B, Sigge GO, Kerwath SE, Hoffman LC (2016) Heavy metals in marine fish meat and consumer health: a review. J Sci Food Agric 96: 32-48.

9. Fakhri Y, Saha N, Miri A, Baghaei M, Roomiani L, et al. (2018) Metal concentrations in fillet and gill of parrotfish (Scarus ghobban) from the Persian Gulf and implications for human health. Food Chem Toxicol 118: 348-354.

10. González RR, Martínez JCL, Milán EG, Frenich AG, Vidal JLM (2007) Simultaneous determination of selected veterinary antibiotics in gilthead seabream (*Sparus aurata*) by liquid chromatography-mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci 857: 142-148.

11. Bajer T, Šilha D, Ventura K, Bajerová P (2017) Composition and antimicrobial activity of the essential oil, distilled aromatic water and herbal infusion from Epilobium parviflorum Schreb. Ind Crops Prod 100: 95-105.

12. Cabello FC (2003) Antibiotics and aquaculture. An analysis of their potential impact upon the environment, human and animal health in Chile. Fundacion Terram. Analisis de Politicas Publicas 1-16.

13. McDermott PF, Zhao S, Wagner DD, Simjee S, Walker RD, et al. (2002) The food safety perspective of antibiotic resistance. Anim Biotechnol 13: 71-84.

14. Liang Y, Tse MF, Young L, Wong MH (2007) Distribution patterns of polycyclic aromatic hydrocarbons (PAHs) in the sediments and fish at Mai Po Marshes Nature Reserve, Hong Kong. Water Res 41: 1303-1311.

15. IPCS (International Programme on Chemical Safety) (2010) Polycyclic aromatic hydrocarbons, selected nonheterocyclic.

16. Kanaki (2005) Analytical methods for the determination of PAHs in sediments, PhD proposal personal communication, University of the Aegean, Department of Environmental Studies Mytilene.

17. Retnam A, Zakaria MP, Juahir H, Aris AZ, Zali MA, et al. (2013) Chemometric techniques in distribution, characterisation and source apportionment of polycyclic aromatic hydrocarbons (PAHS) in aquaculture sediments in Malaysia. Mar Pollut Bull 69: 55-66.

18. Anderson KE, Kadlubar FF, Kulldorff M, Harnack L, Gross M, et al. (2005) Dietary intake of heterocyclic amines and benzo(a)pyrene: associations with pancreatic cancer. Cancer Epidemiol Biomarkers Prev 14: 2261-2265.

19. Hogsette JA, Koehler PG (1991) Pesticide safety around animals. Virginia Cooperative Extension Service (USA).

20. Muir D, Sverko E (2006) Analytical methods for PCBs and organochlorine pesticides in environmental monitoring and surveillance: a critical appraisal. Anal Bioanal Chem 386: 769-789.

21. Mansour SA (2004) Pesticide exposure--Egyptian scene. Toxicology 198: 91-115.

22. Kaur R, Kaur S, Lata M (2011) Evaluation of DNA damage in agricultural workers exposed to pesticides using single cell gel electrophoresis (comet) assay. Indian J Hum Genet 17: 179-187.

23. Taranger GL, Carrillo M, Schulz RW, Fontaine P, Zanuy S, et al. (2010) Control of puberty in farmed fish. Gen Comp Endocrinol 165: 483-515.

24. Abo-Al-Ela HG (2018) Hormones and fish monosex farming: A spotlight on immunity. Fish Shellfish Immunol 72: 23-30.

25. Ibrahim HAM (2009) Hormonal residues in chicken carcasses. Ph.D. thesis, Beni Suef. University Faculty of Vet. Medicine.

26. Henderson BE, Feigelson HS (2000) Hormonal carcinogenesis. Carcinogenesis 21: 427-433.

27. Al-Bahry SN, Mahmoud IY, Al-Belushi KIA, Elshafie AE, Al-Harthy A, et al. (2009) Coastal sewage discharge and its impact on fish with reference to antibiotic resistant enteric bacteria and enteric pathogens as bio-indicators of pollution. Chemosphere 77: 1534-1539.

28. Adedeji OB, Adebisi T, Emikpe B (2017) Bacteria load on the skin and stomach of Clarias Gariepinus and Oreochromis Niloticus from Ibadan, South West Nigeria: Public health implications. J Microbiol Biotechnol Res 1: 52-59.

29. Holden AJ, Chen L, Shaw IC (2001) Thermal stability of organophosphorus pesticide triazophos and its relevance in the assessment of risk to the consumer of triazophos residues in food. J Agric Food Chem 49: 103-106.

30. Farag MM (2013) Effect of different cooking methods on nucleic acid nitrogen bases content of fresh sardine fish and its nutritive value. World Journal of Dairy & Food Sciences 8: 156-164.

31. Abd-Allah, Shimaa S (2013) Studies on chemical and microbiological contaminants in some fish species and the influence of some cooking and processing methods on these contaminants. M. Sc. Thesis, Fac. of Agric. AL-Azher Univ, Egypt.

32. Zorraquino MA, Althaus RL, Roca M, Molina MP (2011) Heat treatment effects on the antimicrobial activity of macrolide and lincosamide antibiotics in milk. J Food Prot 74: 311-315.

33. El-Sherif SA, Ibrahim SM, Abd El-Gahfour SA (2016) The validity of some dominant fish in Wadi El Rayan protected area Lakes, Fayoum, Egypt. J Union Arab Biol. Cairo 23: 1-8.

34. AOAC (2005) Official methods of analysis of the association of official analytical chemists international. 18th edition, Maryland, USA.

35. Bloxham MJ, Gachanja A, Hill SJ, Worsfold PJ (1996) Determination of mercury species in sea-water by liquid chromatography with inductively coupled plasma mass spectrometric detection. J Anal At Spectrom, 11: 145-148.

36. Shin D, Kang H, Jeong J, Kim J, Choe WJ, et al. (2018) Multi-residue Determination of Veterinary Drugs in Fishery Products Using Liquid Chromatography-Tandem Mass Spectrometry. Food Anal Methods 11: 1851-1831.

37. Almatari MS, Yousry MA, Laila R, Naglaa L, Tawfic AM (2017) Residues of some organic pollutants, their bioaccumulation, and risk assessments profile in Lake Temsah, Ismailia, Egypt. J Clin Exp Tox 1.

38. Molina-Ruiz JM, Cieslik E, Cieslik I, Walkowska I (2015) Determination of pesticide residues in fish tissues by modified QuEChERS method and dual-d-SPE clean-up coupled to gas chromatography-mass spectrometry. Environ Sci Pollut Res Int 22: 369-378.

39. Seo J, Kim HY, Chung BC, Hong J (2005) Simultaneous determination of anabolic steroids and synthetic hormones in meat by freezing-lipid filtration, solid-phase extraction and gas chromatography-mass spectrometry. J Chromatogr A 1067: 303-309.

40. Cheung KC, Leung HM, Wong MH (2008) Metal concentrations of common freshwater and marine fish from the Pearl River Delta, south China. Arch Environ Contam Toxicol 54: 705-715.

41. Onsanit S, Ke C, Wang X, Wang K, Wang W (2010) Trace elements in two marine fish cultured in fish cages in Fujian province, China. Environ Pollut 158: 1334-1342.

42. Wang Y, Chen P, Cui R, Si W, Zhang Y, et al. (2010) Heavy metal concentrations in water, sediment, and tissues of two fish species (Triplohysa pappenheimi, Gobio hwanghensis) from the Lanzhou section of the Yellow River, China. Environ Monit Assess 165: 97-102.

43. Ismail I, Saleh IM (2012) Analysis of Heavy Metals in Water and Fish (*Tilapia sp*.) Samples from Tasik Mutiara, Puchong. The Malaysian Analytical Sciences Society 16: 346-352.

44. Türkmen M, Türkmen A, Tepe Y, Ateş A, Gökkuş K (2008) Determination of metal contaminations in sea foods from Marmara, Aegean and Mediterranean seas: Twelve fish species. Food Chem 108: 794-800.

45. Morshdy A, Darwaish S, Hussein M, El Ebidy O (2015). Heavy Metal Residues in Marketed Fish at Manzalla Dakahllyia. 2nd Conference of Food Safety, Suez Canal University, Faculty of Veterinary Medicine. Volume I August: 111-118.

46. Choi J, Park S, Kim YH, Oh SC, Lim ES, et al. (2016) Pathogenic microorganisms, heavy metals, and antibiotic residues in seven Korean freshwater aquaculture species. Food Sci Biotechnol 25: 1469-1476.

47. Done HY, Halden RU (2015) Reconnaissance of 47 antibiotics and associated microbial risks in seafood sold in the United States. J Hazard Mater 282: 10-17.

48. Barani A, Fallah AA (2015) Occurrence of tetracyclines, sulfonamides, fluoroquinolones and florfenicol in farmed rainbow trout in Iran. Food Agric Immunol 26: 420-429.

49. Darko G, Akoto O, Oppong C (2008) Persistent organochlorine pesticide residues in fish, sediments and water from Lake Bosomtwi, Ghana. Chemosphere 72: 21-24.

50. Yamashita N, Urushigawaa Y, Masunaga S, Walash MI, Miyazaki A (2000) Organochlorine Pesticides in Water, Sediment and Fish from the Nile River and Manzala Lake in Egypt. Int J Environ Anal Chem 77: 289-303.

51. Nazli B, Colak H, Aydin A, Hanpikyan H (2005) The presence of some Anabolin Residues in Meat and Meat products sold in Istan. Turk J Vet Animsci 29: 691-699.

52. Suvanich V, Jahncke ML, Marshall DL (2008) Changes in Selected Chemical Quality Characteristics of Channel Catfish Frame Mince During Chill and Frozen Storage. J Food Sci 65: 24-29.

53. EOS (1990) Canned sardine fish. Egyptian Organization for Standardization. Ind Mins, Cairo 287.

54. Shen L, Yang L, Peng T (1996) Amperometric Determination of Fish Freshness by a Hypoxanthine Biosensor. J Sci Food Agric 70: 298-302.

55. Mohamed FSA, Sabae SZ (2015) Monitoring of pollution in Wadi El-Rayan Lakes and its impact on fish. Int J Dev 4: 1-28.

56. Perelló G, Martí-Cid R, Llobet JM, Domingo JL (2008) Effects of various cooking processes on the concentrations of arsenic, cadmium, mercury, and lead in foods. J Agric Food Chem 56: 11262-11269.

57. Ersoy B, Yanar Y, Küçükgülmez A, Çelik M (2006) Effects of four cooking methods on the heavy metal concentrations of sea bass fillets (Dicentrarchus labrax Linne, 1785). Food Chem 99: 748-751.

58. FAO/WHO (1999) Food Agriculture Organization/World Health Organization, Expert Committee on Food Additives. Summary and conclusions of the sixty-first meeting of the Joint FAO/WHO Expert Committee on Food Additives (JECFA) Rome: 22.

59. Uno K, Aoki T, Kleechaya W, Tanasomwang V, Ruangpan L (2006) Pharmacokinetics of oxytetracycline in black tiger shrimp, Penaeus monodon, and the effect of cooking on the residues. Aquaculture 254: 24-31.

60. Hsieh MK, Shyu CL, Liao JW, Franje CA, Huang YJ, et al. (2011) Correlation analysis of heat stability of veterinary antibiotics by structural degradation, changes in antimicrobial activity and genotoxicity. Vet Med (Praha) 56: 274-285.

61. Amal ME, Amani MS (2012) Effect of different traditional cooking styles on pesticides residues in fresh Tilapia nilotica and Clarias lazera fish in Gharbia markets. Alexandria J Veterinary Sci 35: 197-209.

62. Hassanen FS, Kamel EA, Gaafar RAM, Shaheen AA (2016) Effect of Grilling on Pesticides Residues in O. niloticus Muscles. Benha Vet Med J 30: 348-357.

63. Domingo JL (2011) Influence of cooking processes on the concentrations of toxic metals and various organic environmental pollutants in food: a review of the published literature. Crit Rev Food Sci Nutr 51: 29-37.