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Assessment of bacterial, heavy metals and pesticides pollutants in Nile tilapia and catfish

A thesis presented by

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Abstract

(Keywords: Nile tilapia, Catfish, Heavy metals, Pesticides, Microbial hazards, Fry, Grill).

A total of 120 samples of Nile tilapia (*Oreochromis niloticus*) and catfish (*Clarias gariepinus*) examined to assess the presence of different pollutants from bacterial, heavy metals and pesticides hazards associated with them from three different locations in Egypt. The results showed a high prevalence for Coliform, Fecal coliform and Staphylococcus counts and *Pseudomonas aeruginosa* was the dominant microorganism in the examined fish samples, only lead and cadmium were exceeded the national and international permissible limits. The dominant pesticides that were detected in the study were Hexachlorobenzene (HCB), Methyl parathion and Cypermethrin were detected in all fish samples from the three locations. Frying caused significant losses in the concentrations of most of examined heavy metals and pesticides than grilling.

Dedication

To MY Family
Especially My Daughter Maya

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It is a pleasure to record my great deepest thanks to GOD.

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List of Abbreviations

Abbreviations	Name
APC	Aerobic Plate Count
ATSDR	Agency for Toxic Substances and Disease Registry
BHC	Benzene Hexa Chloride
CAC	Codex Alimentarius Commission
Cd	Cadmium
CFU/g	Colony Forming Unit Per Gram
Cr	Chromium
CRFUS	Code of Federal Regulations of the United States of
Cu	America
DDT	Copper
E.coli	Dichlorodiphenyltrichloroethane
EHEC	Escherichia coli
EOSQC	Enterohaemorrhagic E.coli
ESS	Egyptian Organization for Standardization and Quality
EU	Control
FAO	Egyptian Standard Specification
FDA	European Commission Regulation
Fe	Food and Agriculture Organization
Fig	Food and Drug Administration
HCB	Iron
HCH	Figure
IARC	Hexa Chloro Benzene
ICMSF	Hexa Chloro Hexane
	International Agency For Research on Cancer
mg/Kg	International Commission on Microbiological

Mn	Specifications for Food
MPN	Milligram per kilogram
ND	Manganese
Ng/g	Most Probable Number
NK cells	Not Detected
No	Nanogram per gram
OCP	Natural Killer cells
OP	Number
PAHS	Organochlorine Pesticides
Pb	Organophosphorus Pesticides
QuEChERS	Poly Aromatic Hydrocarbons
VTEC	Lead
WHO	Quick Easy Cheap Effective rugged and Safe pesticide
Zn	multi-residue method
	Verocytotoxin E.coli
	World Health Organization
	Zinc

Introduction

INTRODUCTION

In recent decades, food safety has become a widespread public concern worldwide due to the rising demand for eating safe food; this has stimulated research work regarding certain threats associated with food stuffs contaminated with heavy metals, pesticides and microbial toxins. As some investigations have confirmed that more than 90% of human contaminants come from food.

The River Nile is the principle fresh water resource and life for Egypt representing more than 97% of Egypt's water resources (Ali *et al.*, 2008). as a sequence of increasing industry, agriculture, Urbanization and tourism, human activities are responsible for different pollution sources for the environment and the aquatic ecosystems (Bin-Dohaish *et al.*, 2008).

Fish is an important source of food and represents a main part of many natural food chains. Fish is highly nutritious due to its high protein content and the presence of omega-3 fatty acids, fats, amino acids and vitamins; it also contains a number of minerals, including Ca, Fe, Cd, Pb, Cu, and Zn (Safia, 2005; Aremu and Ekunode, 2008). It may carry many contaminants that cause many potential effects on fish itself and the organisms that consume them, including humans (Burger and Gochfeld, 2005).

Nile tilapia is the most popular freshwater fish species in Egypt. It plays an important role in fish consumption in Egypt and around the world this attributed to many positive qualities including tolerance to poor water quality, lie in a wide range of temperature, dissolved oxygen, salinity, pH, light intensity and photo periods. High degree of plasticity in growth and their reproductive biology is characterized by short generation time and show little susceptibility to diseases.

Nile catfish is highly consumed in Egypt because it is low-priced and available in most localities. The African catfish tend to live in turbid and cloudy waters. It is more exposed to different sources of environmental pollutants than other fish types. Because of the relatively high fat content of catfish meat, the fat soluble environmental pollutants such as organochlorine pesticides (OCs) is the more probable pollutants present in catfish meat (Holtan, 1998). Furthermore, the organophosphorus pesticides (OPs) could pollute the tilapia and catfish meat via the recent agricultural application.

The most serious fish safety issues associated with microbial especially bacterial pathogens and organic pollutants with heavy metals and pesticides, so that fish can be used as indicator for environmental pollution for its ability to concentrate pollutants in their tissues (Fisk *et al.*, 2001).

The dangers accompanied with human pathogenic bacteria in fish can be divided into two groups: bacteria naturally present in the aquatic environment, referred to as indigenous bacteria and those present as a result of contamination with human or animal faeces (Reilley, 1998). These pathogenic bacteria are *Vibrio* spp, *Aeromonas* spp, *Salmonella* spp, pathogenic *E. coli*, *Shigella* spp and *Y. enterocolitica*.

Fecal coliform in fish demonstrates the level of pollution of their environment because coliforms are not the normal flora of bacteria in fish (Chon and Shuval, 1973) the enteric bacilli include *E. coli*, *Klebsilla* spp, *Citrobacter* spp, *Enterobacter* spp, *serratia* spp.

In recent years worldwide attention has been paid to the problems of environmental contamination by metals (Malik *et al.*, 2010; Aktar *et al.*, 2011; Qadir and Malik, 2011; Maceda-Veiga *et al.*, 2012). Metals were of particular concern due to their toxicity and ability to bio-accumulate in aquatic ecosystems (Mohammadi *et al.*, 2011), as well as persistency in the natural environment. Among the metals some are potentially toxic (As, Cd, Pb, Hg,

etc.), others are probably essential (Ni, V, Co) and many are essential (Cu, Zn, Fe, Mn) (Biswas *et al.*, 2011). These essential metals can also produce toxic effects when the metal intake is excessively elevated (Tüzen, 2003; Tekin-Özan, 2008).

Heavy metals often enter the environment through human activities, persist in food webs and accumulate in living organisms. In addition to atmospheric sources (Batty *et al.*, 1996), other possible metal sources (Cu, Cd, Pb and Zn) are close to rice fields. Non-essential heavy metals are usually powerful toxins and their bioaccumulation in tissues leads to intoxication, decreased fertility, cellular and of a variety of organs (Oliveira Ribeiro *et al.*, 2000, 2002; Damek-Proprawa and Sawicka-Kapusta, 2003). Essential metals such as copper (Cu), magnesium (Mg), manganese (Mn) and zinc (Zn) have normal physiological regulatory functions (Hogstrand and Haux, 2001), but may also bioaccumulate and reach toxic levels (Rietzler *et al.*, 2001). Effects on the environment of these trace metals, mainly from human discharges are not well identified and more information is necessary to better understand the importance of these pollutants, especially in protected areas.

Fish being at the top level of the food chain that accumulates large quantities of heavy metals in fish depends up on the intake and the removal from the body (Karadede *et al.*, 2004). Heavy metals concentration in various species depends on the feeding behavior (Romeo *et al.*, 1999), size and length of fish (Al-Yousuf *et al.*, 2000) and principally on their habitat (Conli and Atli, 2003).

According to FAO (2002), a pesticide is any chemical that kills controls, repel, or modifies the behavior of a pest. Pesticides can be classified according to their chemical structure to organochlorines, organophosphorus and pyrethroids.

Organochlorine pesticides (OCPs) are synthetic, non polar, toxic and can withstand in the environment for long periods. They are ubiquitous and persistent pollutants due to bioaccumulation in the food chain either as such or as their metabolites, thus causing concern on the animals at the top of the food chain. Due to their persistency and lipophilicity, OCPs from the water column can easily be accumulated by biota. Mostly, fishes are able to uptake contaminants through gills and food intake and, eventually, transfer them to humans through consumption of these organisms (Zhou *et al.*, 2008). Therefore, the accumulation of OCPs in edible tissues is of concern from human health perspective since the fish body load somehow reflects the contamination status of the environment.

The Organophosphorus (OP) insecticides are neurotoxicants are regarded as being less persistent compared with OCs, but some reports have indicated that residues of OPs can withstand for extended periods in organic soil and surrounding drainage systems in Egypt by Abdel-Halim *et al.* (2006). Due to their cheapness and effectiveness to control pests, weeds and diseases (He *et al.*, 2009), they had been widely used and became more and more important in agricultural production after OCs were forbidden. OP residues can concentrate and diffuse by the effect of biological enrichment and food chains; therefore, it might appear in food products and cause potential risk for human health (Sun *et al.*, 2011).

Pyrethroids are synthetic analogues of the natural pyrethroids, extracts of the ornamental Chry-Santemum Cinerariaefolium and its related species used to control of insects for decades, they were selective, safe and had short half lives extremely toxic to fish. Pyrethroids are heat stable and photo stable, slightly soluble in water and highly soluble in fats.

Pesticides reach aquatic environment by direct application, spray drift, aerial spraying and runoff from factories, sewage and agricultural drainage which has been considered a major hazard on human health, (Salah El Dien and Nasr, 2004). They cause severe toxic problems including developmental