

## Effect of Freezing and Different Cooking Methods on Heavy Metals Content of the Nile Tilapia *Oreochromis niloticus*

<sup>1</sup>Nora Ibrahim, <sup>1</sup>Manal El-Garhy, <sup>1</sup>Amr Abdel-Khalek, <sup>1,2</sup>Kareem Morsy and <sup>3</sup>Ali Al Ghamdi

<sup>1</sup>Department of Zoology, Faculty of Science, Cairo University, Cairo, Egypt

<sup>2</sup>Department of Biology, College of Science, King Khalid University, Abha, Saudi Arabia

<sup>3</sup>Department of Biology, College of Science, Al Baha University, Al Baha, Saudi Arabia

**Abstract:** The Nile tilapia (*Oreochromis niloticus*) is a species of tilapia, a cichlid fish native to Africa from Egypt south to east and central Africa; it is one of the most commercially important fish species in Egypt. The present study aimed to evaluate the effects of traditional cooking (Fried and grilled) and freezing processes on the concentrations of Cd, Cu, Fe and Pb in fish fillets of *O. niloticus*. It was observed that in general, the heat treatment by either frying or grilling and freezing of polluted *O. niloticus* fish flesh led to a decrease of the heavy metals content in all the tested fish parts. The decrease of metals which occurred during frying of fish was lower than that detected after the grilling process. Cadmium concentrations (mg/kg dry wt) after the present experiment were: Fresh flesh (0.142) > frozen (0.031) > fried (0.027) > grilled (0.014). For Cu: Fresh flesh (14.905) > frozen (0.936) > fried (0.696) > grilled (0.311). For Fe: Fresh flesh (9.78) > frozen (5.890) > fried (3.085) > grilled (2.998). And for Pb: Fresh flesh (0.058) > fried (0.053) > grilled (0.038) > frozen (0.021). The reduction in the metal contents of the fish during cooking may be related to the release of these metals with the loss of water as free salts, possibly in association with soluble amino acids and coagulated proteins. Therefore, it is possible to reduce the metal in fish parts by choosing a suitable method of cooking. Grilling had a more pronounced effect on the heavy metals content in fish parts than frying.

**Key words:** *Oreochromis Niloticus* • Freezing • Cooking • Fillet • Heavy Metals

### INTRODUCTION

Fish constitute a major source of protein and healthy lipid throughout the world and its consumption has increased globally in the recent time. Fish provides the long-chain polyunsaturated omega-3 fatty acid which might favorably improve lipid profile and reduce cholesterol levels, the risk of coronary heart diseases, stroke and preterm diseases [1-3]. Environmental pollution represents a major problem in both developed and underdeveloped countries. Egypt is one country which suffers from high biosphere pollution (air, soil and water). Many ecological changes occur in water as a result of human activities, including agricultural, industrial and municipal wastes [4]. Cadmium (Cd), copper (Cu), lead (Pb) and zinc (Zn) salts are usually found in agricultural and industrial liquid wastes [5-8] which are discharged into water resources. These metals are toxic to aquatic life at

low concentrations, particularly in soft-water environments [9]. Such metals may be accumulated from water to higher levels in fish tissues [10]. Evidently these metals accumulate frequently in fish flesh [11-14] and internal organs [15-18]. Many elements, which are present in sea food, are essential for human life at low concentrations but they can be toxic at high concentrations. Therefore, many consumers regard any presence of these elements in fish as a hazard to health [19]. The heavy metals concentrations of fish fillets can be affected by processing or cooking methods and therefore, it is important to determine the concentrations of heavy metals in raw and cooked fish fillets and it is possible to reduce the heavy metal concentration in fish fillets by choosing a suitable method of cooking [20-22]. Therefore, it is important to determine the concentrations of essential and nonessential metals in raw and cooking fish in order to evaluate the possible risks of fish consumption for

human health [23]. Therefore, the main purpose of the present study was to evaluate the effects of traditional cooking (Fried, grilled) and freezing processes on the concentrations of Cd, Cu, Fe and Pb in various foods.

## MATERIALS AND METHODS

**Fish Collection and Preparation of Fillets:** Fresh fish samples were collected from a location along the River Nile at Rokn Farouk, Helwan, Giza governorate, Egypt during summer 2017. Fifty fresh fish samples Nile Tilapia (*Oreochromis niloticus*), mean length and weight were 18.45 cm and 105 g; were captured transported alive to the ecology laboratory, Zoology department, Faculty of Science, Cairo university, Egypt and then they were re-washed thoroughly with potable water then beheaded, gutted and again washed to get their fillets. The obtained fish fillets were washed with tap water to remove any traces of blood and viscera. After that, first part fish fillets immersed in 10% sodium chloride solution for 10 min, then washed with tap water and treated with spices and finally fillets were battered using edible batter which was prepared according to Abdou *et al.* [24] by mixing 94% wheat flour, 2% egg yolk, 2% skimmed milk, 1.8% salt and 0.2% cumin with water by 1:3 (w: w) and this ingredients were homogenized for 2 min. After the batter coating, it was covered with bread crumbs. After that, every fish species fillets were divided into four batches, the first batch was control (without coating and without cooking process), the second was stored as frozen. The third batch was cooked grilling by microwave oven (Samsung, model at 980 watt) for 5 min each side of fish fillets samples. The fourth batch was cooked frying using Halogen oven (Lentel, model KYR-912A, 1300C watt) heated at 180°C for 5 min of each side of fish fillets samples. All cooked fish fillets samples were allowed to cool at ambient temperature and packaged in polyethylene bags and stored until analysis.

**Heavy Metals Analysis:** Control, cooked as well as frozen fillets were prepared for heavy metals analysis according to the method described by Meche *et al.* [25] as follows: fish fillets were dried in an oven at 105°C over night. The samples were removed from the oven, allowed to cool and ground in a clean mortar and pestle. Approximately 0.5 g of each sample was placed in a Teflon microwave digestion bomb with 10 mL of concentrated HNO<sub>3</sub>. The samples were allowed to ramp to 180°C for 5 min, digest at 180 °C for 9.5 min and cool down for 5 min in microwave digestion system (Model Milestone, MLS-1200

mega, Germany). The samples were then transferred to clean volumetric flasks and diluted with H<sub>2</sub>O to 10 ml. The samples were stored at 5°C until ready for analysis of metals. The metals Cd, Cu, Fe and Pb were determined using inductively coupled plasma atomic emission spectrometry (Varian model- Liberty Series II) against aqueous standards (mg/kg dry wt). All metal concentrations were expressed as mg/kg dry wt.

**Statistical Analysis:** The results were expressed as means ± S.D. Data were statistically analyzed using Duncan's multiple range test to evaluate difference in means as indicated by different case letters at P<0.05. The statistical analysis was done using SAS version 9.1.

## RESULTS AND DISCUSSION

The concentrations of heavy metals with means and standard deviation are given Table (1). It was observed that in general, the heat treatment by either frying or grilling and freezing of polluted *O. niloticus* fish flesh led to a decrease of the heavy metal content in all the tested fish parts. The decrease of metals which occurred during frying of fish was lower than that detected after the grilling process. Cadmium concentrations after the present experiment were: Fresh flesh (0.142) > frozen (0.031) > fried (0.027) > grilled (0.014). For Cu: Fresh flesh (14.905) > frozen (0.936) > fried (0.696) > grilled (0.311). For Fe: Fresh flesh (9.781) > frozen (5.890) > fried (3.085) > grilled (2.998). And for Pb: Fresh flesh (0.058) > fried (0.053) > grilled (0.038) > frozen (0.021).

Concentrations of heavy metals in different fish species showed great variation capabilities for accumulating heavy metals. The observed variability of heavy metals levels in different species depend on, feeding habits [26] ecological needs, metabolism [27] age, size and length of the fish and their habitats [28] species, sex, biological cycle and on the part of the fish analyzed [29]. Factors such as season, location, environment of development, nutrient availability and temperature and salinity of the water, may contribute to variations in the metal concentrations in fishes [3]. It was interesting to note that, in most cases, fish fillets showed significant decrease in heavy metal concentrations after cooking by microwave and halogen. Similar results were recorded by Atta *et al.* [20] who found a significantly decrease in the concentrations of Mn, Pb and Zn in Nile tilapia after cooking methods. However, Gokoglu *et al.* [30] found a significant effect on mineral content due to cooking. Moreover Ersoy *et al.* [21] reported that, the heavy metals

Table 1: Mineral composition (mg/kg dry wt) of fresh, cooked and frozen fillets samples of *O. niloticus* showed considerable effect on reducing trace metals concentrations in fish (Data were expressed as mean ±SD).

<i>Oreochromis niloticus</i>				
	Fresh	Frozen	Fried	Grilled
Cd	0.142±0.041 <sup>a</sup>	0.031±0.003 <sup>b</sup>	0.027±0.003 <sup>b</sup>	0.014±0.004 <sup>b</sup>
Cu	14.905±1.136 <sup>a</sup>	0.936±0.060 <sup>b</sup>	0.696±0.051 <sup>b</sup>	0.311±0.019 <sup>b</sup>
Fe	9.781±1.143 <sup>a</sup>	5.890±1.737 <sup>ab</sup>	3.085±1.225 <sup>b</sup>	2.998±0.318 <sup>b</sup>
Pb	0.058±0.003 <sup>a</sup>	0.021±0.006 <sup>b</sup>	0.053±0.004 <sup>ac</sup>	0.038±0.003 <sup>c</sup>

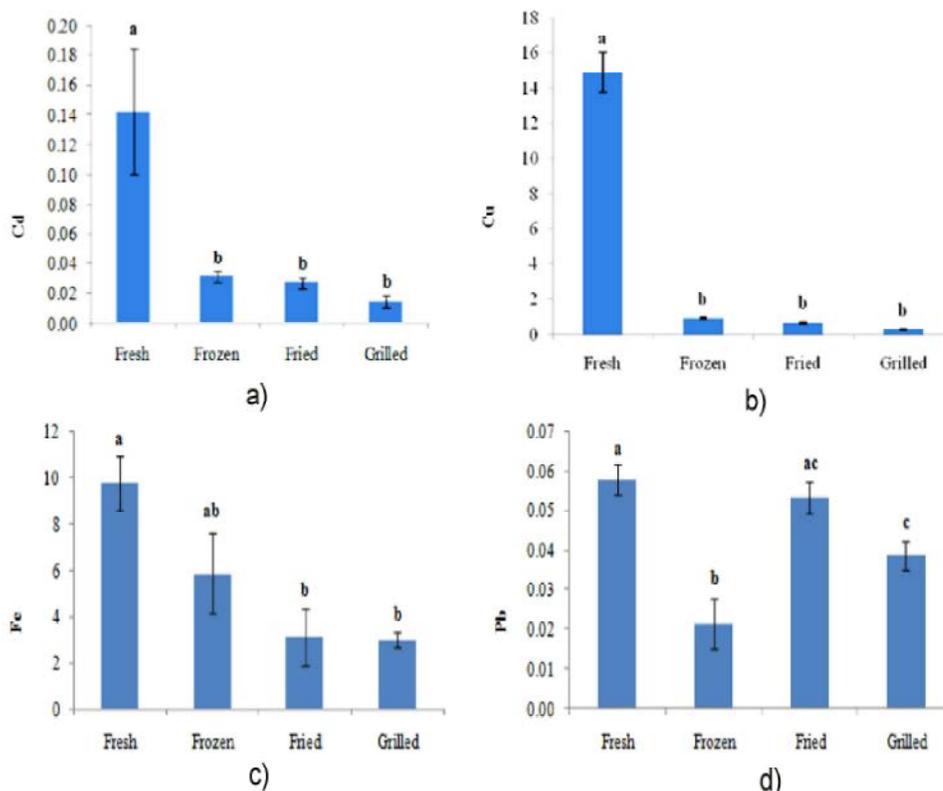


Fig. 1: Bioaccumulated heavy metals (mg/kg dry wt) in the muscle tissue of *O. niloticus* fillets samples as fresh, frozen, fried and grilled; a) Cd, b) Cu, c) Fe, d) Pb

content in all fish parts decreased after baking. Similarly, Hassanin [31] found a significant decrease in Pb content of fillets after cooking by baking and microwave oven methods. The reduction in Pb depends on cooking conditions, such as time, temperature and medium of cooking. Also, Baking, microwaving and grilling showed a more pronounced effect on the heavy metal content [22] while frying lead to a moderate decrease in heavy metals load during the cooking process. The reduction in trace metals concentrations as affected by cooking methods may be due to the release of these metals with the loss of drip as free salts, possibly in association with soluble amino acids and un-coagulated proteins bounded with metals [20, 21, 32]. In conclusion,

Concentrations of heavy metals in different fish species showed great variation capabilities for accumulating heavy metals. The reduction in the metal contents of the fish during cooking may be related to the release of these metals with the loss of water as free salts, possibly in association with soluble amino acids and coagulated proteins [33]. The cooking process decreased the protein content of the fish parts [34]. Hence, heavy metals usually bind with proteins [35]. Cd, Cu, Fe and Pb metals are predominantly accumulated in fish viscera, head and flesh tissues. Fish viscera had the highest contents of heavy metals followed by the fish head. The lowest values were detected in fish flesh. Cooking has effect on the means of reducing the

concentrations of heavy metals and the reduction depends on cooking conditions (Time, temperature and medium of cooking) [36-39]. Therefore, it is possible to reduce the metal in fish parts by choosing a suitable method of cooking. Grilling had a more pronounced effect on the heavy metals content in fish parts than frying. Although fish flesh had the lowest contents of heavy metals, it lost a moderate amount of these during cooking. Consequently, such fish flesh should only be eaten after cooking. Modern cooking techniques for grilling by microwave oven

#### ACKNOWLEDGMENT

The authors would like to express their gratitude to the Faculty of Science, Cairo University, Cairo, Egypt for providing administrative and technical support.

#### REFERENCES

1. Burger, J. and M. Gochfeld, 2005. Heavy metals in commercial fish in New Jersey. *Environ. Res.*, 99: 403-412.
2. Musaiger, A. O. and R. D'Souza, 2008. The effects of different methods of cooking on proximate, mineral and heavy metal composition of fish and shrimps consumed in the Arabian Gulf. *Arch. Latinoam. Nutr.*, 58:103-109.
3. Bashir, F.A., M. Shuhaim-Othman and A.G. Mazlan, 2012. Evaluation of trace metal levels in tissues of two commercial fish species in Kapar and Mersing coastal waters, Peninsular Malaysia. *J. Environ. Public Health* 2012:352309. Article ID352309, 10 pages, doi:10.1155/2012/352309.
4. Katz, M., G.L. Pederson, M. Yoshinaka and D. Sjolseth, 1969. Water pollution (Effect of pollution on fish life). *J.W.P.C.F.*, 41: 9941015.
5. USEPA, 1975. United State Environmental Protection Agency. Scientific and technical assessment report on cadmium EPA-600/6-75003, Washington, DC, USA.
6. Simon, R.D., 1977. Public health implication of enzymes hydrolysis fish protein concentration and waste water based aquaculture. M.Sc. Thesis, University of California, Berkeley, CA, USA.
7. McCrea, R.C. and J.D. Fischer, 1986. Heavy metal and organochlorine contaminants in the five major Ontario Rivers of the Hudson bay lowland. *Water Poll. Res. J. Can.*, 21: 225-234.
8. El-Gamal, I.M., 1993. An environmental evaluation of industrial wastewater as a source of irrigation. *Proc. 3<sup>th</sup> International Conf on Environ. Protect. Is a Must*, April, 13- 15, Alexandria, Egypt, pp: 207-219.
9. USEPA, 1977. United State Environmental Protection Agency. Quality criteria for water. Office of Water and Hazardous Materials, Washington, DC, USA.
10. Forstner, U. and G.T. Wittman, 1983. *Metal Pollution in the Aquatic Environment*, 2nd edn. Springer, New York, pp: 486.
11. Causeret, J., 1962. Fish as a source of mineral nutrition. In: *Fish as Food*, Vol. 11, ed. Borgstrom, pp: 205-228.
12. Jaakkola, T., H. Takahashi and J. Miettinen, 1973. Cadmium content in sea water, bottom sediment, fish, lichen and elk in Finland. In *Environmental Quality and Safety, Global Aspects of Chemistry, Toxicology and Technology as Applied to the Environment*, eds. F. Coulston and F. Korte. Academic Press, New York, pp: 230-333.
13. Underwood, E.J., 1977. *Trace Elements in Human and Animal Nutrition*. Academic Press, New York, pp: 196-242.
14. Glover, J.W., 1979. Concentration of arsenic, selenium and ten heavy metals in school shark, *Galeorhinus australis* (Macleay) and gummy shark, *Mustelus antarcticus* Gunther, from south eastern Australian waters. *Aust. J. Mar. Fresh. Water Res.*, 30: 505.
15. Venugopal, B. and T. Luckey, 1975. Toxicity of non-radioactive heavy metals and their salts. In *Heavy Metal Toxicity, Safety and Hormology*, ed. F. Coulston. Academic Press, George Thieme, Stuttgart, New York.
16. Saianki, J., V.B. Katahne and B. Erzsebet, 1982. Heavy metals in animals of Lake Balaton. *Water Res.*, 16: 1147-1152.
17. Nishihara, T., T. Shimamoto, K.C. Wen and M. Kondo, 1985. Accumulation of lead, cadmium and chromium in several organs and tissues of carp. *J. Hygienic Chem.*, 31: 119-123.
18. Wageman, R., 1989. Comparison of heavy metals in two groups of ringed seals (*Phocahispide*) from the Canadian Arctic. *Can. J. Fish. Aquat. Sci.*, 46: 125.
19. Oehlenschlager, J., 2002. Identifying heavy metals in fish. In H. A. Bremmer (Ed.). *Safety and quality issues in fish processing* (Vol. 507, pp. 95-113). USA: CRC Press LLC.

20. Atta, M.B., L.A. Sabaie, M.A. Noaman and H.E. Kassab, 1997. The effect of cooking on the concentration of heavy metals in fish (*Tilapia nilotica*). Food Chemistry, 58: 1-4.
21. Ersoy, B., Y. Yanar, A. Kucukgulmez and M. Çelik, 2006. Effects of four cooking methods on the heavy metal concentrations of the sea bass fillets (*Dicentrarchus labrax* Linne, 1785). Food Chem, 99: 748-751.
22. Diaconescu, C., G.L. Fantanaru, L. Urdes, Vidu B. Vasile and S. Diaconescu, 2013. Influence of cooking methods over the heavy metal and lipid content of fish meat. Rom. Biotech. Lett., 18(3): 8279-8283.
23. Cid, B.P., C. Boia, L. Pombo and E. Rebelo, 2001. Determination of trace metals in fish species of the Ria de Aveiro (Portugal) by electro-thermal atomic absorption spectrometry. Food Chemistry, 75: 93-100.
24. Abdou, E.S., A.S. Nagy Osheba and M.A. Sorour, 2012. Effect of chitosan and chitosan-nanoparticles as active coating on microbiological characteristics of fish fingers. Int. J. Appl. Sci. Tech., 2(7): 158-169.
25. Meche, A., M.C. Martins, E. S. Bruna, N. Lofrano, Carey J. Hardaway, M. Merchant and L. Verdade, 2010. Determination of heavy metals by inductively coupled plasma-optical emission spectrometry in fish from the Piracicaba River in Southern. Brazil Micro chem. J., 94: 171-174.
26. Romeo, M., Y. Siau, Z. Sidoumou and M. Gnassia-Barelli, 1999. Heavy metal distribution in different fish species from the Mauritania coast. Sci. Total Environ, 232: 169-175.
27. Canli, M. and R.W. Furness, 1993. Toxicity of heavy metals dissolved in sea water and influences of sex and size on metal accumulation and tissue distribution in the Norway lobster *Nephrops norvegicus*. Mar. Environ. Res., 36: 217-236.
28. Canli, M. and G. Atli, 2003. The relationships between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species. Environ. Pollut., 121(1): 129-136.
29. Tuzen, M., 2003. Determination of heavy metals in fish samples of the middle Black Sea (Turkey) by graphite furnace atomic absorption spectrometry. Food Chem., 80(1): 119-123.
30. Gokoglu, N., P. Yerlikaya and E. Cengiz, 2003. Effects of cooking methods on the proximate composition and mineral contents of rainbow trout (*Oncorhynchus mykiss*). Food Chem., 84: 19-22.
31. Hassanin, S.I.A., 2008. Metals residues, histological alterations and cooking methods of fish cultured in wastewater ponds. Proceeding of the 8<sup>th</sup> International Symposium on Tilapia in Aquaculture, pp: 569-588.
32. Ganbi, H.H.A., 2010. Heavy metals pollution level in marine Hammour fish and the effect of popular cooking methods and freezing process on these pollutants. World J. Dairy Food Sci., 5(2): 119-126.
33. Bryan, G.W. and L.G. Hummerstone, 1971. Adoption of the polychaete. nereisdiversicolor to sediments containing high concentrations of heavy metals. J. Mar. Biol. Ass. UK, 51: 845-863.
34. Howarth, R.S. and J.B. Sprague, 1978. Copper lethality to rainbow Trout in waters of various hardness and pH. Water. Res., 12: 455-462.
35. Manar, M.A.F., 2013. Effect of different cooking methods on nucleic acid nitrogen bases content of fresh Sardine fish and its nutritive value. World J. Dairy Food Sci., 8(2): 156-164.
36. Nessrien, M.N.Y. and A.T. Mohamed, 2007. Antioxidant and Antimicrobial effects of marjoram and thyme in coated refrigerated semi fried Mullet fish fillets. World J. Dairy Food Sci., 2(1): 01-09.
37. Hend, H.A.G., 2010. Heavy metals pollution level in marine Hammour Fish and the effect of popular cooking methods and freezing process on these pollutants. World J. Dairy Food Sci., 5(2): 119-126.
38. Mousumi, A., M.D.J. Islam, M. Sohel, H.S. Fatema, H.R. Mohammad and M.D. Kamal, 2014. Changes in Fillet Quality of Pangas Catfish (*Pangasianodon hypophthalmus*) During Frozen Storage. World J. Marine Sci., 6(2): 146-155.
39. E.E. Adebote and M. Eke, 2018. Survey on problems and solutions on method of production and processing of fish in delta state. World J. Marine Sci., 10(2): 18-23.