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Full Length Research Paper

Assessment of industrial waste from the Nile tilapia (*Oreochromis niloticus*) processing industry concerning trace metals and contaminants

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Abstract

The fish business growth in Brazil has raised concerns about sustainability of the sector; the large amount of waste generated from this activity has become a serious problem. This study aims to perform, for the first time in Brazil, the chemical and environmental assessment of industrial waste of Nile tilapia (*Oreochromis niloticus*) processing industry regarding occurrence of toxic components and risks to the environment, human and animal health. Twenty-four waste samples obtained from eight different days from fish processing were evaluated. The procedures to collect samples and aliquots followed the standards of ABNT-NBR 10007:2004 and the methods used followed the recommendations of Standard Methods for the Examination of Water and Wastewater APHA/AWWA/WEF (1995) and ABNT standards. All samples were classified as Class II, type "non-inert" because of the levels of lead, manganese, surfactants and iron. Fish waste, if disposed incorrectly, directly into water resources without proper treatment, can cause risks to the environment, human and animal health, since heavy metals could be leached. Therefore, the waste classification before re-use or final treatment is necessary. In this case, the waste could be used as raw material to produce value-added by-products, more specifically for energy production. This procedure would earn to this fish processing industry a designation of eco-efficient.

Keywords: waste management, industry waste, sustainability

Introduction

Waste is any material discarded in production processes, which, due to technological or market limitations, does not have economic value, requiring technically and economically viable solutions for its management (Brasil, 2010).

The processing of fish products requires and consumes a large amount of water and generates a high amount of waste, which should be added to the waste generated at capturing and marketing (Islam *et al.*, 2004; Kuca and Szaniawska, 2009; Saidi *et al*., 2014; Drost *et al*., 2014; Gullu *et al*., 2015).

The fish industry waste is the non-usable fraction of fish, due to market and/or technological limitations, but that contains chemical characteristics similar to the raw material (Dragnes *et al*., 2009). Annually, approximately 30 million tons of fish waste are discarded worldwide. Most waste is not handled properly, resulting in a waste of raw materials and causing negative environmental, economic and social impacts (Arvanitoyannis and Kassaveti, 2008; Hi-Kittikun *et al.*, 2012; FAO, 2014a; Haider *et al.,* 2016).

In 2011, tilapia cultivation worldwide amounted to approximately 3.95 million tons and for 2030, production expected to growth by 30% (FAO, 2014b). Tilapia accounted for 41% of aquaculture production in Brazil in

2013, and, from 2007 to 2011, it grew from 95,091 tons to 253,824.1 tons, showing an increase of 266% (Brasil, 2011; Brasil, 2013).

The volume of waste generated during tilapia processing is significant and indicates the need to develop appropriate management plans for the sector. In the process to obtain fresh or frozen fillets of tilapias, the average yield is approximately 30%, resulting in about 70% of waste including head, carcass, viscera, skin and scales (Shirahigue *et al.,* 2014,Valente *et al.,* 2014).

In Brazil, fish waste is disposed into water bodies nearby production places (Stori*et al.,* 2002; Spillere; Beumord, 2006; Valente *et al.,* 2014). This material, rich in organic matter, with the presence of large amounts of nitrogen and minerals such as calcium and phosphorus, can lead to eutrophication and accumulation of nitrogen metabolites. (Galvao *et al.,* 2009; *Chowdhury et al.,* 2010; Molisani *et al.,* 2013). The disposal of these effluents into water environments (Sipaúba-Tabi *et al.,* 2008; Manetti *et al.,* 2011) could damage water quality worsening water stress and scarcity, for example, the water crisis that recently hit the state of São Paulo (Agência Nacional de Águas - ANA, 2014).

Increased monitoring, under sanctions to "paying-polluter" after the regulation of the Brazilian Solid Waste Policy – PNRS (Brazil, 2010), has valued low environmental impact, animal welfare, presence of environmental labelling and waste recovery of the productive process (Brasil Food Trends 2020, 2010; Pieniak *et al.,*2013).

The deployment of PNRS marks the beginning of a legal framework that was non-existent until then. It is believed that academic research should encompass aspects of PNRS for the construction of scientific papers on this theme (Cesar *et al.,* 2015). In order to define the best alternatives for management of fish industry waste, it is crucial to characterize and classify the waste according to Brazilian standards, allowing to formulate actions that are possible to be implemented as well as estimate environmental and public health risks.

The alternative to recover, treat and dispose fish solid waste depends directly on its chemical composition, the levels of contaminants and its physical state (Ribeiro *et al.,* 2011). ABNT "NBR 10004 – Solid Waste – Classification" was drafted in 1987 and revised in 2004 by the Brazilian Association of Technical Standards (ABNT), aiming at classifying solid waste regarding its danger, considering its potential risks to the environment and public health, to ensure proper management. However, waste from the production of animal-origin food is not included in Lists 1 and 2 of this standard. There are no data available showing the classification of solid wastes from tilapia processing in accordance with annexes F and G of ABNT-NBR 10004 (Brazilian Association of Technical Standards – ABNT, 2004). It is crucial to obtain greater knowledge about the characteristics and risks associated with fish industry waste to define appropriate actions aimed at minimizing, recovery, treatment and proper disposal of fish waste. This study performed, for the first time in Brazil, the chemical and environmental evaluation of industry waste of Nile tilapia, the main species produced worldwide.

Material and Methods

Sampling – tilapias and waste

Twenty-four samples were collected in eight processing days of tilapias along the course of a year. In each processing day, 75 fish, weighing about 500 g, were processed (22°12'38"S/49°39'22"W). The total waste of each processing day was fully ground to allow homogenization using Metvisa industrial equipment (model CUT and 2.5). From the homogenized mixture, three samples of 3 kg each were collected.

Waste characterization and classification

For the chemical-environmental analysis and classification of waste, the samples were analyzed in triplicate, according to Brazilian Association of Technical Standards (ABNT – NBR, 2004) to characterize and classify the waste. The procedure of removing aliquots for the analyses followed the Standards ABNT – NBR 10007:2004 and the methods used in the chemical analyses followed the recommendations of the "Standard Methods for the Examination of Water and Wastewater" (APHA/AWWA/WEF, 1995) and ABNT Standards (NBR 10004, 2004; NBR 10005, 2004; NBR 10006, 2004).

Risk assessment

To evaluate risks and determine the presence of possible contaminants, a survey was held at the fish production farm used in the tilapia processing. This information allowed the identification of the waste origin, selection of physical and chemical parameters to be analyzed and consultation of Lists 1 and 2 of ABNT – NBR 10004 (ABNT, 2004). The leaching test was performed according to ABNT – NBR 10005:2004 methodology.

Solubility analysis

After obtaining the soluble extract, as recommended by ABNT-NBR 10006 (2004), the pH was measured by the EPA method 9045c (Environmental Protection Agency - EPA, 1996).

Analyses of heavy metals and contaminants

To determine the chemical parameters in the soluble and leachate extracts, the samples were subjected to the preliminary treatment that consisted of digestion with HNO_3 (50 mL extract + 5 mL acid), following the EPA method 3015 (EPA, 1996), using the technique of microwave digestion in closed system (CEM, 2000).

The levels of metals in digested extracts were determined in atomic absorption spectrophotometer Varian-Zeeman (Model 640-Z), equipped with graphite furnace and cold steam generator (EAA–FG).

The analyses were conducted according to the Standard Methods for the Examination of Water and Wastewater (APHA/USEPA – SW 846, 1995). The method of methylene blue active substances (5540 C method) was used in the analysis of surfactants. Chlorides and nitrates were analyzed by colorimetry (4500-Cl-B, and 4500-NO3 methods). The sulphides were determined by the turbidimetric method (4500-SO42- E method). After the calculation of the means and standard deviation, the results were compared with Annex G of norm ABNT-NBR 10004 (ABNT, 2004).

Statistical analysis

The data were subjected to the analysis of variance according to appropriate model to complete randomized design and subsequently for means comparisons in the Tukey test with predetermined significance level at 5% ($p < 0.05$). The analyses were carried out using the SAS statistical computing system (SAS, 2002).

Results

Animal waste from food industrial production is not included in Lists 1 and 2 of ABNT-NBR 10004 (2004).
In order to characterize the hazardous traits of the samples, the leaching test was carried out according to ABNT-NBR 10005:2004 methodology and the sample results were compared to the thresholds in Annex F of ABNT-NBR 10004 (2004).

Table 1 shows the pH values, concentration of metals and contaminants (mg/L) in the leached extract of filleting waste of Nile tilapia (*Oreochromis niloticus*) within the period from August 2007 to August 2008.

Table 1: pH values, concentration of metals and contaminants (mg /L) in the leached extract of filleting residue of Nile Tilapia (*Oreochromis niloticus*)

Parameter	Aug/07	Sep/07	Oct/07	Dec/07	Feb/08	May/08	Jul/08	Aug/08	ML***
pH	5,71± 0,32	5,87± 0,26	5,89±0,39	5,75±0,45	6,20±0,21	5,69±0,38	5,70±0,43	5,75±0,41	2– 12
As	< 0,30 **	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	1,00
Ba	0,54 ± 0,10	0,58 ± 0,05	0,61 ± 0,07	0,63 ± 0,09	0,57 ± 0,13	0,49 ± 0,06	0,52 ± 0,10	0, 59 ± 0,08	70,00
Cd	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	< 0,30	0,50
Pb	0,07±0,01	0,06±0,02	0,07±0,01	0,06±0,01	0,06±0,02	0,07±0,04	0,08±0,03	0,07±0,02	1,00
Cr	0,59 ± 0,08	0,5 ± 0,10	0,52 ± 0,07	0,49 ± 0,12	0,51 ± 0,10	0,6 ± 0,07	0,6 ± 0,1	0,58 ± 0,07	5,00
Mg	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	0,10
Ag	<0,30	<0,30	<0,30	<0,30	<0,30	<0,30	<0,30	<0,30	5,00
Se	0,03 ±0,01	0,02 ±0,01	0,03 ±0,01	0,03 ±0,01	0,03 ±0,01	0,02±0,01	0,02±0,02	0,03±0,01	1,00

All values did not differ statistically at 5% in the Tukey test;
** Values after the symbol (<) mean that the results were lower than the detection limits of the methodology.
*** ML: Ceilings as per Annex F da ABNT - NBR 10.004 (2004) (mg/L).
Note: Annex F is not restricted to the pollutants mentioned in this table.

The values of Table 1 are below the thresholds in Annex F of ABNT-NBR 10004 (2004).
To classify class II waste, as inert or non-inert, the procedure of obtaining soluble extract of solid waste and the comparison of the sample results with the thresholds in Annex G of ABNT-NBR 10004 (2004) was used.

Table 2 shows the concentration of metals and contaminants (mg/L) in the soluble extract of filleting waste of Nile tilapia (*Oreochromis niloticus*).

Table 2: Concentration of metals and contaminants (mg / L) in the solubilized extract of filleting residue of Nile Tilapia (*Oreochromis niloticus*)

Parameter	Aug/07	Sep/07	Oct/07	Dec/07	Feb/08	May/08	Jul/08	Aug/08	ML
Al	0,02±0,01	0,02±0,01	0,02±0,02	0,02±0,01	0,02±0,01	0,02±0,01	0,02±0,01	0,02±0,02	0,20
As	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	0,01
Bário	0,33±0,04	0,32±0,03	0,33±0,06	0,32±0,02	0,34±0,03	0,33±0,05	0,33±0,05	0,34±0,04	0,70
Cd	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	0,005
Hg	0,05±0,01	0,04±0,03	0,06±0,03	0,05±0,04	0,05±0,02	0,06±0,04	0,05±0,03	0,05±0,02	0,01
Cr	0,05±0,03	0,05±0,04	0,04±0,03	0,05±0,01	0,05±0,03	0,06±0,01	0,06±0,07	0,05±0,02	0,05
Cu	1,96±0,10	1,97±0,09	1,92±0,13	1,95±0,07	1,93±0,04	1,94±0,10	1,95±0,08	1,96±0,07	2,00
Mn	52,26±0,17	54,78±0,22	59,56 ±0,34	56,62±0,25	54,68±0,32	55,32±0,19	52,45±0,24	53,37±0,24	0,10
Ag	0,05±0,01	0,04±0,03	0,05±0,03	0,02±0,03	0,03±0,02	0,04±0,04	0,05±0,03	0,05±0,02	0,05
Se	0,0021±0,0	0,0020±0,01	0,0024±0,03	0,0020±0,01	0,0021±0,03	0,0021±0,03	0,0020±0,02	0,0018±0,03	0,01
Na	125,25±1,23	127,13±2,34	124,45±1,88	125,76±0,94	133,62±0,79	131,32±1,97	127,39±1,71	125,25±1,2	200,00
Zn	2,23±0,24	2,34±0,45	2,89±0,74	3,01±0,64	3,11±0,62	2,76±0,47	2,67±0,31	2,27±0,42	5,00
Cloretes	198,63±1,16	197,43±1,32	198,89±1,87	195,83±2,34	196,34±1,70	194,99±2,12	197,84±1,12	198,12±1,13	250,00
Nitrates	8,00±0,10	7,89±0,21	8,06±0,29	8,02±0,18	8,32±0,20	8,09±0,10	7,94±0,16	8,01±0,15	10,00
So_{4-}	175,36±2,15	171,48±1,87	173,89±1,34	175,75±1,94	174,83±1,84	175,29±0,78	171,67±1,58	174,55±0,42	250,00
Surfactants	1,04±0,11	1,00±0,23	1,05±0,18	1,07±0,28	0,98±0,12	0,93±0,51	0,99±0,26	1,03±0,27	0,50
Fe	55,69±2,41	54,89±3,47	52,33±3,68	53,91±1,96	55,12±1,86	59,21±3,64	58,21±1,34	56,31±2,03	0,30
Mg	<0,001**	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	0,001

All values did not differ statistically at 5% in the Tukey test;
** Values after the symbol (<) mean that the results were lower than the detection limits of the methodology.
*** ML: Ceilings as per Annex G da ABNT - NBR 10.004 (2004) (mg/L).
Note: Annex G is not restricted to the pollutants mentioned in this table.

The concentrations of lead, manganese, iron and surfactants in the soluble extract were higher than levels established in Annex G of ABNT-NBR 10004 (2004) (Table 2).

The concentration of metals and contaminants in the samples showed no significant difference (P > 0.05) during the collection period (Tables 1 and 2).

Discussion

With respect to origin, production and conditions of fish, it was not observed in the production site and its vicinity, the practice of using antibiotics and agrochemicals that could have contaminated the cultivation sites and, consequently, the fish. Although organic contaminants Aldrin, cyanide, chlordane, 2.4 D, DDT, Endrin, heptachlor, hexachlorobenzene, lindane and methoxychlor are listed in annexes G and H of ABNT-NBR 10004 (ABNT, 2004), they were not detected in the samples analyzed in this study.

The pH values of samples of fish industry wastes did not present significant differences throughout the collection period and point to a nonhazardous trait of material (Table 1). The maximum values found at 6.2 are within the range recommended by the Sanitary and Industrial Inspection of Products of Animal Origin (Brasil, 2017) to the inside of fresh fish (< 6.5) and lower than the threshold recommended by ABNT-NBR 10004 (ABNT, 2004).

The results obtained in the leached extract (Table 1) were below the threshold in Annex F of ABNT-NBR 10004 (ABNT, 2004). In addition, due to their known origin and collection characteristics, the samples do not have features of corrosiveness, inflammability and pathogenicity. Therefore, the samples should not be regarded as hazardous waste confirming Alfonso *et al.,* (2004).

However, concentrations of lead, manganese, iron and surfactants in the soluble extract were higher than the thresholds in Annex G of ABNT-NBR 10004 (ABNT, 2004) (Table 2). Thus, all samples of industry waste of tilapias (*Oreochromis niloticus*) analyzed are classified as class II – waste, “non-inert type”. The concentrations of manganese, lead, surfactants and iron were, respectively, 530, 5, 2 and 186 times greater than the maximum allowed (Table 2) and did not suffer influence of the environmental conditions during the collection period.

Silver and chrome values were within the limits established by ABNT-NBR 10004 (ABNT, 2004) and copper levels were too close to the maximum value. Although some values of metals are not yet at critical levels today, they can be dangerous in the future (Amoo *et al.,* 2005; Silva *et al.,* , 2015); therefore, limits of copper and silver and other metals should be monitored.

The concentration of surfactants is attributed to the use of sanitizers and water agents during the industrialization. Iron concentration, in turn, can be attributed to its solubility in water and be influenced by interactions with other soluble constituents. In addition, iron participates in liver processes and is also associated with the hemoglobin transport. Iron is considered one of the most important elements for the respiratory process. Therefore, iron concentrations in the waste samples analyzed may be attributed to the occurrence of iron, mainly in fish guts, as a blood constituent. Also, the concentrations may have originated in sulfates and chlorides of iron and magnesium, commonly used in water treatment processes during fish processing (Braile and Cavalcante ;1993; Birungi *et al.,* 2007; Silva *et al.,* 2015).

Among the parameters that showed concentrations higher than the thresholds of ABNT-NBR 10004 (ABNT, 2004) for soluble extracts, manganese and lead stand out, as these metals are considered neurotoxic for being able to induce neural dysfunction or cause damage to the peripheral system (Sissino, 2003). The manganese tendency to precipitation in oxides in water significantly reduces its availability to aquatic organisms and, consequently, the possibility to cause direct toxic effects (Arenzon and Raya-Rodriguez, 2006).

The lead values may have been originated from bio-accumulative processes due to local contamination of crops, which may originate in domestic sewage near the cultivation sites (Molisani *et al.,* 2013). For Carvalho *et al.* (2000), lead is toxic to fish thus there is a need for monitoring environmental conditions and zootechnical parameters.

The finding of high levels of these metals in the industry waste of tilapias highlights the need for proper monitoring and disposal of this waste material, considering mainly contamination of groundwater through solubilization and leaching as well as the bio-accumulative and toxic effects of these elements (Amoo *et al.,* 2005; Tiping and Lofts, 2013).

The Nile tilapia (*Oreochromis niloticus*) is one of the most common species of freshwater used in toxicological studies and is an indicator in biomonitoring programs (Eneji *et al.,* 2011; Manarino *et al.,* 2011). Because they are omnivorous, tilapias feed all tracks of the water column. The viscera of these fish are used for analyses of metals due to the bioaccumulation process. Therefore, the metals may have accumulated in fish guts corroborating the results presented in this study (Birungi, *et al.* 2007; Abdel-Baki *et al.*, 2011; Canpolat, 2013).

The waste disposal into water bodies or in places where there may be leaching of metals and organic matter can affect the autochthonous or cultivated flora and fauna (Cui, 2011; Manetti *et al.,* 2011). Contaminants may be transported by water over long distances. The path traversed by these contaminants depend on their stability and physical state and water flow (Mwandya *et al*., 2010; Manarino *et al.,* 2011).

In case metals are leached and solubilized in the waste and disposed into the environment, the standards of effluent disposal established by the National Council for the Environment – CONAMA (Brasil, 2005) showed that manganese with 54.88 mg/L (average value for samples) and iron with 55.70 mg/L (average value for samples) were above the threshold determined by the law, respectively, 1 and 15 mg/L.

Fish waste is characterized mainly by the organic matter content, a potential source of bacterial decay. If the common practice of disposing waste material directly into the soil or into water environments, besides reducing the oxygen concentration in water, increasing biochemical demand for oxygen, it can accumulate or export the metals in the samples from these locations, causing risks to the environment and public health (Catchpole and Gray, 2010; Chowdhury *et al.,* 2010; Suuronen and Sarda, 2007; Molisani *et al.,* 2013).

Studies show that contamination by metals can cause a wide variety of clinical exposures ranging from motor disorders and behavioral changes to psychosis. Research on plants and animals of environments affected by waste disposal or industrial effluents provide evidence of genotoxic effects and neoplasia (Passos and Mergler, 2008; Nyland *et al.,* 2011; Stankovic *et al.,* 2014).

Therefore, inadequate management of this waste can impair the quality of water, soil and fish cultured nearby, and if waste is disposed near urban areas, it could compromise the entire ecosystem (Torres de Oliveira *et al.,* 2013). Characterization and classification of this solid waste should be conducted to guide the special care in management to obtain new products and ensure safety to public health and/or the environment (Ribeiro *et al.,* 2011, Campos and Galiza, 2016).

The industrial waste of Nile tilapia processing industry can be classified as class II (non-inert), offers potential for use in by-products thus reinstated into the economic cycle. Therefore, the waste classification before re-use or final treatment is necessary. Several alternatives have been developed with this purpose using various types of fish waste (Bourtoom *et al.,* 2009; Ferraz de Arruda *et al.,* 2009; H-Kittikun *et al.,* , 2012; Centenaro *et al.,* 2014; Shirahigue *et al.,* 2014; Santos *et al.,* 2015, Gullu *et al.,* 2015). However, due to concentrations of lead, manganese, surfactants and iron found in the samples this fish waste should be used to obtain new by-products that are not aimed directly at food consumption.

The waste material could be used for energy generation as an alternative to fuel or as a motor cell (Vázquez and Murado, 2008; Shimoyama *et al.,* 2009; Wiggers *et al*. 2009; Chowdhury *et al.,* 2010; Wisniewsky Junior *et al.,* 2010; Kramer *et al.,* 2011; Souza *et al.,* 2012, Santos et al*, 2015*).

Conclusion

The industrial waste from the Nile tilapia (*Oreochromis niloticus*) processing industry could be classified as Class II (non-inert) in terms of levels of lead, manganese, surfactants and iron, since the concentrations of these pollutants were above the threshold recommended by the ABNT.

When disposed inadequately, which is a reality for industry fish processing in Brazil, metals can be leached, causing risks to the environment, human and animal health.

As was observed in this study, the chemical-environmental assessment is a measure necessary for the implementation of appropriate management systems on the part of industry and regulatory agencies to prevent environmental pollution, which may occur and be aggravated in the short-term by the increasing production of tilapias and to indicate which co-products can be obtained from the use of these materials.

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