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Efficiency of the anaerobic baffled reactor followed by anaerobic filter in the removal of nutrients and pathogenic organisms in fish processing effluents

[*Eficiência do reator ABR seguido de filtro anaeróbio na remoção de nutrientes e de organismos patogênicos em efluentes de processamento de pescado*]

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ABSTRACT

The aquaculture showed high growth along with the increase in the consumption of animal protein from this sector. The processing industries facilitate the preparation of fish for the consumer; however, they generate large volumes of effluents with a high polluting potential. Environmental legislation establishes norms for the release of effluents, making it necessary to implement treatment systems to reduce the pollutants generated. The objective of this work was to evaluate the performance of a compartmentalized anaerobic reactor (ABR) followed by an anaerobic filter (AF) treating fish processing effluent. The work was carried out in a slaughterhouse that had an effluent treatment station consisting of a static sieve, grease box, ABR reactor and anaerobic filter. Monitoring consisted of physical-chemical and biological analyzes of samples collected from the influent and effluents from each stage of treatment. The parameters evaluated were ammonia, nitrite, nitrate, NTK, phosphate and coliforms. The average results of the removal efficiency of these parameters, respectively, for the ABR reactor were 5, 40, 69, -19, -25 and 83%, and for the AF -0.5, 73, 53, 10, -17 and -17%. The system composed by the ABR reactor followed by the Anaerobic Filter showed high removal of nitrite, nitrate, and coliforms.

Keyword: agroindustry, environmental impact, anaerobic digestion

RESUMO

A atividade de aquicultura apresentou elevado crescimento, juntamente com o aumento do consumo de proteína animal proveniente desse setor. As indústrias de processamento facilitam o preparo do pescado ao consumidor, todavia geram grandes volumes de efluentes de alto potencial poluidor. A legislação ambiental estabelece normas para o lançamento de efluentes, tornando necessária a implementação de sistemas de tratamento para a redução dos poluentes gerados. O objetivo deste trabalho foi avaliar o desempenho de um reator anaeróbio compartimentado (ABR) seguido por filtro anaeróbio (FA), tratando efluente de processamento de pescado. O trabalho foi desenvolvido em um frigorífico que possuía uma estação de tratamento de efluentes composta por peneira estática, caixa de gordura, reator ABR e filtro anaeróbio. O monitoramento consistiu em análises físico-químicas e biológicas de amostras coletadas do afluente e dos efluentes de cada etapa do tratamento. Os parâmetros avaliados foram: amônia, nitrito, nitrato, NTK, fosfato e coliformes. Os resultados médios da eficiência de remoção desses parâmetros, respectivamente, do reator ABR foram de 5, 40, 69, -19, -25 e 83%, e do FA -0,5, 73, 53, 10, -1, e -17%. O sistema composto pelo reator ABR seguido pelo filtro anaeróbio apresentou alta remoção de nitrito, nitrato e coliformes.

Palavras-chave: agroindústria, impacto ambiental, digestão anaerobia

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INTRODUCTION

The production of aquatic organisms, called aquaculture, has become one of the fastest growing food activities in the world (Zhao *et al*., 2021). Fish is the main source of animal protein in the human diet and a large part of the aquaculture sector's feed is made up of proteins (Nissa *et al*., 2021).

Due to world population growth, the demand for fish production has been growing (Nissa *et al*., 2021), making the fish processing market interesting for industries, such as ways of preparing fish for the final consumer. It was estimated for the year 2018 a global production of fish around 179 million tons, with 156 million tons of this total destined for human consumption (The state…, 2020).

Fish processing industries generate large amounts of solid and liquid waste. Most fish processing industries carry out common activities such as filleting, drying, freezing, fermenting, canning, and smoking. In these steps, processing effluents are generated, which can contain organic matter in soluble, colloidal, and particulate forms. However, the quantities of these effluents generated, and the pollutant loads can vary according to the type of process carried out and according to each industrial unit (Palenzuela-Rollon, 1999; Chowdhury *et al.*, 2010). For example, Balslev-Oslev *et al*. (1990) described total chemical oxygen demand (COD) values for herring processing wastewater for brine production of 90 g L^{-1}; Panpong *et al*. (2014) reported COD value for wastewater from the processing of canned seafood of 10.4g L^{-1}; Jemli *et al*. (2015) presented total organic carbon (TOC) values for fish processing wastewater of 11.5 g L^{-1}; Sanjaya *et al*. (2020) reported COD values for fish processing wastewater around 30 to 35 g L^{-1}. Therefore, the values of organic material contained in effluents for different types of fish processing vary widely.

In the fish processing industries, the large amount of effluent generated results from the high use of water in practically all stages, including cleaning the raw material, cooking, cooling, washing floors and cleaning equipment (Cristóvão *et al*., 2015). The inadequate disposal of this type of effluent in soils and, mainly, in bodies of water without adequate treatment, can cause serious environmental damage, bringing biological risks to both humans and living organisms in the affected ecosystem (Sankpal and Naikwade, 2012; Santos *et al*., 2022).

The effluent from fish processing has a high load of organic nutrients, coming mainly from carbonaceous compounds and nitrogen. In addition, it can also contain dissolved and suspended solids and the presence of microorganisms (Ching and Redzwan, 2017). However, accessing information on the characterization of this type of effluent for the elaboration of projects of effluent treatment systems on a full scale is difficult due to the great variability in the concentrations of contaminants in the effluents, which can be associated with the design of the processing plants, the wide variation in existing fish species, variations in water consumption demands during processing and in the production process schedules (Jamieson *et al*., 2017). Furthermore, there is still little information on the characterization of effluents resulting from the processing of Nile tilapia, in addition to information on the treatment of fish effluents in anaerobic reactors. Thus, knowledge about the treatment of this type of industrial waste and the environmentally adequate final destination is extremely important and should be considered for the practice of a sustainable activity.

Anaerobic digestion can be used as an alternative for the treatment of fish processing effluents to reduce their polluting potential. The compartmentalized anaerobic reactor (ABR) resembles an improved septic tank, with a series of baffles that are used to direct wastewater in an upward flow to subsequent compartments, thus allowing greater contact between the effluent to be treated and the microorganisms present in the sludge reactor (Barber and Stuckey, 1999; Pirsaheb *et al*., 2015). Among the advantages of applying anaerobic digestion in ABR reactors, we can mention simple design and low cost, possibility of maintaining low hydraulic retention times (HRT) and high solids retention time, considerable stability to organic load shocks and low generation of slime (Barber and Stuckey, 1999). Anaerobic treatment may not be high enough to reach acceptable standardized values for effluent discharge, mainly for organic matter parameters (BOD and COD) and nutrients. However, one of the technological solutions is the combination of different systems, such as the use of an ABR reactor followed by an Anaerobic Filter (AF) (Yosefi *et al*., 2018).

At the federal level, the environmental legislation promoted by the National Environmental Council called 'CONAMA Resolution Nº 430/2011', establishes norms for the conditions and standards for the discharge of effluents into receiving water bodies, making it necessary to search for alternatives for reduction of contaminants generated in activities with potential pollutants. The standard release pattern for the release of effluents into water bodies is a tool that, together with the quality standard of receiving bodies, aims to protect the quality of water sources and the preservation of aquatic ecosystems.

Thus, the management of waste from potentially polluting activities, carried out in an appropriate and efficient manner, is extremely important. In addition, information on effluents from the processing of Nile tilapia (*O. niloticus*) species can guide entrepreneurs in the field to implement effluent treatment systems as an environmental control measure, to preserve the environment and to meet the environmental requirements and standards determined by current legislation. In this sense, the aim of this study was to evaluate the performance of an ABR reactor followed by a full-scale anaerobic filter, treating effluents from a fish slaughterhouse.

MATERIAL AND METHODS

This study was carried out in a fish slaughterhouse located in the rural area of the southern region of Minas Gerais, on the banks of the Furnas reservoir located within the Rio Grande hydrographic basin. The slaughterhouse had a complete production cycle of Nile tilapia (*Oreochromis niloticus*), performing fish reproduction, larviculture, fattening and fish processing. The fish slaughterhouse processed around 1.2 tons of tilapia per day.

The effluent treatment station (ETS) (Fig. 1) was composed of a static sieve, grease box, ABR and an anaerobic filter (AF) in full scale. The grease box, equalization tank, ABR reactor and anaerobic filter (AF) had useful volumes of 2.9L, 54.7L, 100.8L and 50.9 L, respectively. The ETS was designed to process six fish metric tons per day, and a waiting discharge of 150m³.day^{-1}. The ascension speed of the system was 0.8m.h^{-1}. The effluent was conducted by the gravity to all treatment stages through tubes and connections. The hydraulic retention time (HRT) used to the ABR (Tab. 1) was about 2.2 days, with organic loading rate (OLR) about 0.5 g chemical oxygen demand (COD) (L.d)$^{-1}$. The HRT for the anaerobic filter was about 1.1 day with OLR about 0.4 g COD (L.d)$^{-1}$.

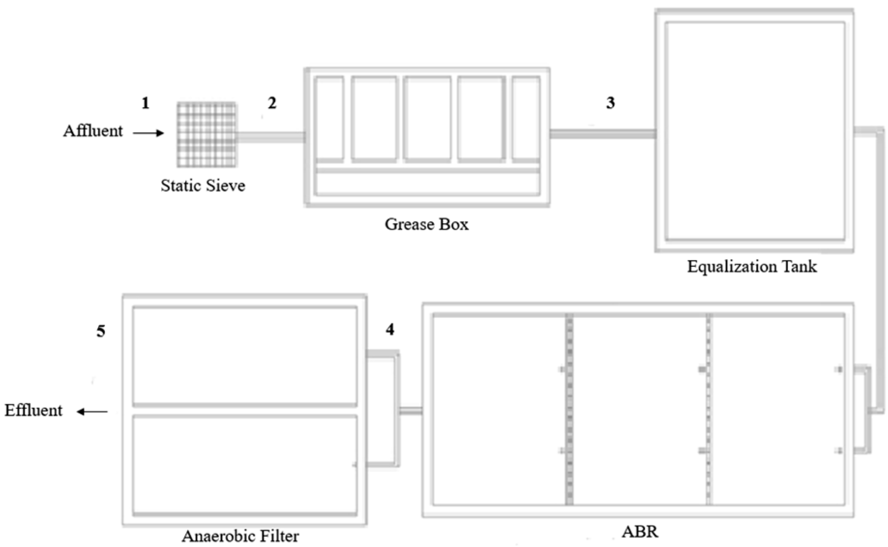

Figure 1. Schematic drawing of the wastewater treatment system.

The monitoring of the ETS consisted of a set of physical-chemical and biological analyzes of the influent and effluent of each stage of treatment. The treatment system was monitored for a period of 10 months. To evaluate the performance of the system composed by the ABR-AF reactors samplings of the influent and effluents were carried out fortnightly and analyzed at the Sanitation Laboratory of the School of Veterinary Medicine at the Federal University of Minas Gerais (UFMG).

To measure the pH of the samples, a bench pH meter was used (Hi 2221 Calibration Check pH/ORP Meter). The total chemical oxygen demand (COD) (method 5220 D), total Kjeldal nitrogen (TKN) (method: 4500-NO_3), ammonia (method: 4500-NH_3 E), nitrite (method: 4500-NO_2 A), nitrate (method: 4500-NO_3 A), phosphates (method: 4500-P E) and thermotolerant coliforms were performed as described in APHA (American…, 2017). For the quantification of thermotolerant coliforms, the Colilert method (method: 9223 B) was used.

Table 1. Operational conditions of the system

ABR				Anaerobic Filter (AF)		
Discharge (L/s)	HRT (day)	COD ($mg.L^{-1}$)	OLR g COD $(L.d)^{-1}$	HRT (day)	COD ($mg.L^{-1}$)	VOL g COD $(L.d)^{-1}$
48	2.2	1320	0.5	1.1	543	0.4

HRT: Hydraulic Retention Time; COD: Chemical Oxygen Demand; VOL: Volumetric Organic Loading

RESULTS AND DISCUSSION

The large generation of effluents from industrial activities raises concerns worldwide. Thus, environmental regulations force industries to apply efficient treatment technologies that are cost-effective and ecologically sound capable of ensuring the ability to manage wastewater sustainably (Kamali *et al*., 2019).

Anaerobic digestion is a biological process that converts biodegradable organic waste into biogas, increasingly attracting the scientific and commercial community, as it is not only about treating organic waste and effluents, but also an alternative that has great potential for renewable energy production and nutrient recycling (Mata-Alvarez *et al*., 2000; Li *et al*., 2014; Chen *et al*., 2015). This waste treatment technology is a very efficient alternative for both the treatment of agricultural and food processing waste, urban and industrial wastewater, and for the treatment of solid waste such as fruit and vegetable waste (Cabezas *et al*., 2015; Mazareli *et al*., 2016).

There are several treatment processes and different ways to use effluents that are highly efficient, but most of these processes require specialized technical assistance and large investments for installation. However, many enterprises and, mainly, small rural producers that work in the agroindustry are not able to use very sophisticated waste treatment methods (Oza *et al*., 2019).

Wastewater from fish processing contains high levels of biodegradable organic material, and due to its characteristic, it has a high potential for treatment through anaerobic processes with possibilities for energy use through the generated biogas. In this case (Palenzuela-Rollon *et al*., 2002). The ABR reactor can be considered a promising anaerobic system technology for the treatment of effluents with high organic loads. The ABR was developed in the 1980s and has great advantages over conventional anaerobic reactors, such as longer biomass retention time, lower energy consumption and greater stability for organic and hydraulic shock loads (Yang *et al*., 2018). However, the combination of treatment systems, such as ABR reactor followed by an anaerobic filter (AF), may be necessary for the treated effluent to reach the standards required by environmental legislation.

Anaerobic filters can be defined as biological reactors filled with materials with good adhesion surface, which can be immobile or inert. This type of system facilitates the proliferation of anaerobic microorganisms, which form a thin layer, called biofilm or slime, surrounding a filler that acts as an absorbent. In addition, they are easy to construct and operate, are considered low cost, and can have high organic material removal efficiencies, generating a low amount of sludge (Souza *et al*., 2010; Tonetti *et al*., 2011; Oza *et al*., 2019).

The average values of the concentrations of ammonia, nitrite, nitrate, TKN and phosphates verified in the influent and effluents of the treatment system composed of an ABR reactor followed by an anaerobic filter are available in Table 2.

It was possible to verify that the concentrations of nitrite, nitrate and TKN reduced from the effluent to the effluent of the system (Table 2). In addition, for these parameters, the determined concentrations will be presented within the conditions and standards for the release of effluents established by Normative Resolution COPAM / CERH nº. 1/2005.

Table 2. Averages of physical and chemical characters obtained at the affluent and effluent of the ABR and Anaerobic Filter

Parameter	Affluent ($mg.L^{-1}$)	ABR ($mg.L^{-1}$)	Anaerobic Filter ($mg.L^{-1}$)
Ammonia	18.8	24.5	24.6
Nitrite	0.0077	0.0088	0.0024
Nitrate	4.5	2.8	1.3
TKN	86.1	35.7	32.3
Phosphates	9.2	9	10.6

TKN: Total Kjeldahl nitrogen

The biological processes which can remove nitrogen in an environment are the nitrification and denitrification. According to Zoppas (2016), the process to promote nitrogen removal involves separate aeration and non-aeration steps. Furthermore, it is necessary that there is an external source of carbon in the denitrification

According to the concentrations of nitrite and nitrate observed in the system, we could infer that there was a coexistence of two processes in the anaerobic filter, namely, nitrification and denitrification. The reduction of concentration of nitrite may be related to the oxygen entry and formation of a biofilm in the aerobic part of the anaerobic filter, which had contact with air and lighting, since the filter was not hermetically closed. Thus, the nitrification may have occurred during this process, converting the ammonia nitrogen into nitrite, and this in nitrate. Concomitantly, the anoxic part of the filter may have allowed the removal of nitrate from its conversion into nitrogen gas by denitrifying bacteria.

In contrast, the increase of the concentration of ammonia and phosphates was recorded in the system. The conversion process of organic nitrogen into ammonia nitrogen is common in anaerobic systems, where the nitrogen is converted into ammonia nitrogen, depending on reduction of pH, due to presence of volatile acids (Chernicharo, 2016). Ammonia nitrogen is an essential nutrient for microbial digestion and contributes to increased alkalinity, being an important ally in anaerobic digestion for the stability of the treatment system (Ariunbaatar *et al*., 2015). The anaerobic digestion is a complex natural process that occurs in stages, in which various intermediate compounds are generated, in addition to methane and carbon dioxide, which are obtained under conditions of microbial association. The interdependence between these microorganisms, however, is the key factor in this process (Singh and Prerna, 2008). One of the advantages of an effluent stabilized in anaerobic digestion is that it can be applied to the soil as a fertilizer in greater safety, as its organic load after the treatment process is reduced (Doll and Foresti, 2010). The ammonia nitrogen and phosphates found in the effluent after treatment are above the limit stated in the environmental law for their discharge in water bodies, requiring the addition of a specific treatment system seeking the nutrients removal. Other alternative to reduce the effluents discharge with high concentration of nutrients in water bodies is to reuse the wastewater, such as in the irrigation, for instance, considering all implication to the public health and attending to standards and requirements for its reuse (Sperling, 2016a).

The overall average removal efficiency is shown in the Table 3, which was about -31% and -15% for ammonia and phosphates, respectively. The negative efficiency for ammonia suggests anaerobic activity. However, the system showed overall average removal efficiency about 69% for nitrite and 71% for nitrate, showing that there was also an aerobic activity.

Table 3. Averages of removal efficiency of the ABR-AF system

Parameter	Removal efficiency (%)		
	ABR	Anaerobic Filter	Overall average
Ammonia	5	-0,5	-31
Nitrite	40	73	69
Nitrate	69	53	71
TKN	-19	10	63
Phosphates	-25	-17	-15

TKN: Total Kjeldahl nitrogen

The negative removal efficiency for ammonia may be related with its production during the biological decomposition process. During the decomposition process of organic matter containing proteins, and during the urea hydrolysis, the organic nitrogen is converted into ammoniac nitrogen. The anaerobic digestion process includes processes such as hydrolysis, acidogenesis, acetogenesis and methanogenesis. The acidogenesis process also generates ammonia nitrogen. The step of acidogenesis, however, consists of a set of fermentative bacteria which convert complex organic compounds such as carbohydrates, proteins, and lipids, in other less complex compounds, by means of hydrolysis and fermentation. During the acidogenesis, occurs the formation of volatile fatty acids, and production of byproducts such as ammonia, carbon dioxide and hydrogen sulfide (Hwang and Hansen, 1998; Pilarska, 2018).

According to Sperling (2016b), anaerobic processes are related to the unsatisfactory removal of nutrients such as nitrogen and phosphorus. Regarding the ABR-based system, this process may result in loss of solids, if occur great variation and excessive picks of influent flow, since the system does not have the auxiliary mechanism for retaining biomass. Although systems of secondary treatment do not show satisfactory nutrients removal for the effluent discharge into water bodies, the negative removal efficiency shown in the system may point out some ETS designing failures. The increase of nutrient concentration, such as phosphorus (Table 2), may be due to drag of sludge in the system. During the monitoring period of the fish slaughterhouse effluent treatment plant, it was verified that the influent velocity and the discharge volume at the entrance of the treatment plant increased in a certain period of the day, when the slaughterhouse was washed and sanitized. Residual waters from fish processing were sent to the gravity treatment system, with no pumping to the ETS to equalize the affluent. Thus, the speed and flow of the effluent at the entrance to the treatment system provided a sludge escape due to the high influent velocity. In this way, the grease box was releasing sedimented material into the system.

The verified values of thermotolerant coliforms for wastewater from fish processing, in this study, was 3.59×10^2 MPN 100mL-¹. These values found were higher than the values reported for fish processing effluent reported by Ferraciolli *et al*. (2017), of $1\text{x}10^3$ MPN 100mL-¹.

The concentration of thermotolerant coliforms at the effluent of the ABR and AF, and the average removal efficiency of treatments are shown in the Table 4. There was a reduction of the thermotolerant coliforms concentration in the refrigerator wastewater (affluent) after treatment in the ABR. According to Sperling (2016a), intestinal origin organisms such as fecal coliforms, among others, show natural mortality when exposed to a different environment from the human body, which is ideal to their reproduction and development. Other factors, such as physical, chemical, biological, and biochemical may also contribute to microbial mortality.

Table 4. Averages of concentration and removal efficiency of thermotolerant coliforms of the ABR and AF system

Averages of concentration of thermotolerant coliforms (MPN 100 mL^{-1})			Averages of removal efficiency of thermotolerant coliforms (%)		
Affluent	ABR effluent	AF effluent	ABR	AF	Overall average
3.59×10^{2}	4.12×10^{1}	4.81×10^{1}	83	-16.71	87

MPN: The most probable number

Although the coliforms removal efficiency in the anaerobic filter (Table 4) did not reach good results, the overall efficiency provided final effluent with low coliform values. Besides factors already described, which may contribute to the coliforms mortality, the reduction of bacterial concentration may have been due to the use of chlorinated water in the fish processing industry. According to Sperling (2016a), the chlorine is one of the most used substances as disinfectants for water and sewage. Its disinfectant action is mainly related to oxidation mechanisms of cellular material. When they are added to water, chlorine compounds react to form hypochlorous acid (HOCl), which decomposes into hypochlorite ion (OCl^-) and hydrogen ion (H^+). The hypochlorous acid has a higher disinfecting power than the OCl^-.

The end result of the concentration of thermotolerant coliforms verified in the effluent of the ETS of the fish slaughterhouse allows the effluent to be reused for irrigation, since it is within the reuse standards established by the World Health Organization, about 1×10^{3} MPN mL^{-1} (Wprld…, 1989).

CONCLUSION

The secondary treatment system of the fish processing plant's ETS monitored in this study, consisting of an ABR reactor followed by an anaerobic filter, demonstrated a high capacity for removing nitrite, nitrate, TKN and thermotolerant coliforms. On the other hand, for the parameters of ammonia and phosphates, the system did not present removal efficiencies, indicating that the increase in the concentrations of these parameters may have resulted from the dragging of solids contained in the sieve and by the dragging of sludge into the grease box. The ideal choice of effluent treatment technologies depends on several factors that include not only the achievement of high efficiencies of compost removal, but also the economic cost, operational criteria, environmental and available information about the residues to be treated. Taking into account that this study has information on the monitoring of a fish processing effluent treatment plant, operated on a full scale, the results obtained are relevant for other industries in the field to apply anaerobic digestion as a form of environmental control, adjusting and adapting each stage of treatment when necessary.

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