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# Employment of chitosan in the removal of pollutants in fish processing effluents

[*Emprego de quitosana na remoção de poluentes em efluentes de processamento de pescado*]

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## ABSTRACT

The significant growth of the industrial sector in recent decades has led to an increase in the volume of waste, which if not properly destined, could cause serious environmental problems. In the context of aquaculture, liquid effluents with a high organic content are generated in large quantities in the fish processing industries, and if their disposal is carried out improperly, serious damage to the environment is caused. The general objective of this study is to evaluate: the efficiency of removal of COD and BOD, in addition to the influence on pH; alkalinity; chlorides; ammonia; nitrite; nitrate; phosphate; turbidity; total, fixed and volatile solids, from the effluent of fish processing treated by coagulation and sedimentation using the natural chitosan coagulant. pH 5.5 followed by pH 6 showed better results for the use of chitosan coagulant in the process of treatment by effluent coagulation of fish processing industry. Chitosan does not act on the parameters TVS, alkalinity, chlorides, ammonia, nitrite, nitrate, and phosphate, regardless of the dosage used. However, it acts on BOD, COD, TS, TFS and turbidity. Thus, the best dosage of chitosan is 0.25 g $L^{-1}$ in optimized activity at pH of 5.5.

Keywords: jar test; water quality, fish effluent, *Oreochromis niloticus*

## RESUMO

*O crescimento expressivo do setor industrial nas últimas décadas acarretou o aumento do volume de resíduos, que, se não forem destinados adequadamente, poderão causar sérios problemas ambientais. No contexto da aquacultura, efluentes líquidos com um alto teor orgânico são gerados em grandes quantidades nas indústrias de processamento de pescado, e, se seu descarte for realizado de maneira inadequada, há sérios prejuízos ao ambiente. O objetivo geral deste estudo foi avaliar: a eficiência de remoção de DQO e DBO, além da influência sobre o pH, a alcalinidade, os cloretos, a amônia, o nitrito, o nitrato, o fosfato, a turbidez, os sólidos totais, fixos e voláteis, bem como do efluente do processamento de pescado tratado por coagulação e da sedimentação, utilizando-se o coagulante natural quitosana. O pH 5,5, seguido pelo pH 6, apresentou melhores resultados para uso do coagulante quitosana no processo de tratamento por coagulação de efluente de indústria de processamento de pescado. A quitosana não atua sobre os parâmetros STV, alcalinidade, cloretos, amônia, nitrito, nitrato e fosfato, independentemente da dosagem utilizada. Mas atua sobre DBO, DQO, ST, STF e turbidez. Assim, a melhor dosagem de quitosana é 0,25 g $L^{-1}$ em atividade otimizada no pH de 5,5.*

*Palavras-chave:* jar-test, *qualidade de água, efluente de pescado,* Oreochromis niloticus

## INTRODUCTION

Aquaculture is an activity that has been increasing significantly in recent decades. According to the report The State of world fisheries and aquaculture (The State…, 2020), in 2016, fish production on the world market by aquaculture activity was 76.5 million tons. In 2017, this production increased to 79.5 million tons, and it continued to grow in 2018 when production reached 82.1 million tons. As

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aquaculture is growing, the concern with the destination/treatment of waste generated by the sector also has increased until the final product was obtained.

Liquid effluents with a high organic content are generated in large quantities in the fish processing industries and, if their disposal is done improperly, they will cause serious damage to the environment. Solid and liquid waste from processing must be separated and the liquid effluent must be subjected to appropriate treatment.

The National Council for the Environment (Conama), through its Resolution No. 430, of May 13, 2011 (Brazil, 2011), provides for the conditions and patterns of effluent release, informing the organic and inorganic parameters to be obeyed by any polluting source.

The treatment of effluents can be carried out by physical-chemical or biological means. The choice of treatment processes considers the type of effluent, its biodegradability, the presence of toxins and the production of sludge, among other factors. Given the ease and low operational cost, the biological process is chosen for the treatment of urban effluents and certain types of industrial effluents (Motta *et al.*, 2003), as is the case of waste from the fish processing industry.

Therefore, the study carried out focuses on the coagulation process, where the main objective, according to Bertoncini (2008) is the removal of solids and organic matter, reducing the values of BOD (biochemical oxygen demand) and DQA (chemical oxygen demand).

In the conventional primary water treatment, aluminum and iron salts are used as a coagulant substance, which at the end of the treatment result in residues with polluting potential. Therefore, alternative natural coagulants are being studied for this purpose (Capelete and Brandão, 2013). The use of natural coagulants has shown advantages over chemical coagulants, such as biodegradability, low toxicity, and low sludge production (Moraes, 2004).

Among the natural coagulants, one currently highlighted is chitosan, a substance extracted from chitin. Chitosan is a natural, low-cost, renewable, and biodegradable product of great economic and environmental importance (Azevedo *et al*., 2007). In addition to its uses in the pharmaceutical and cosmetic industries, several studies are being carried out with the application of chitosan in water treatment due to its coagulant property (Kawamura *et al*., 1993).

Coagulants of natural or synthesized organic origin, universally known as polyelectrolytes, consisting of large molecular chains, are endowed with sites with positive or negative charges, which can, in the presence of water, transform into cationic or anionic coagulants, depending on the equilibrium. electrical charge (Borba, 2001). Being a polyelectrolyte, and due to the high density of positive polymeric charges (Mathur and Narang, 1990), chitosan acts by coagulating organic and inorganic particles, suspended and dissolved, found in water (Capelete and Brandão, 2013).

As the exoskeletons of crustaceans are the main natural sources of chitin, which is a natural fiber precursor of chitosan. The use of chitin as a raw material to produce chitosan would allow the recycling of large amounts of waste from the fishing industry.

In this context, the objective of this study was to evaluate the efficiency of using chitosan under different hydrogen potentials (pH) as a primary treatment for this type of effluent, having the water quality parameters as response variables.

## MATERIAL AND METHODS

The experiment was conducted at the Environmental Sanitation Laboratory of the Department of Preventive Veterinary Medicine - DMVP of the Veterinary School of the Federal University of Minas Gerais - UFMG, located in the municipality of Belo Horizonte, Minas Gerais - Brazil.

The wastewater was obtained from the processing of fish from a slaughterhouse located in the central region of Minas Gerais. The activities developed in this industry were: evisceration and filleting of tilapia (*Oreochromis niloticus*), and the residuary water from these procedures and from the cleaning of surfaces of the processing room was destined to be in a single outlet.

The experimental design was of the multivariate type, using a 3x6 factorial design, with 3 treatments related to pH (T1=5.5; T2=6.0 and T3=6.5) associated with 6 chitosan dosages: 0.25; 0.5; 0.75; 1.0; 1.25 and 1.5 g $L^{-1}$. The whole experiment was performed in duplicate. The results were analyzed by ANOVA, using test of means for the data with the statistical program SISVAR version 5.6.

To simulate the coagulation on bench scale, tests were performed using the microcontrolled, six-test, Labor jar test, with rotation regulator of the mixing rods. The fast mixing (VMR) and slow (VML) speeds employed were 100 rpm and 40 rpm, respectively. The rapid (TMR) and slow (TML) mixing times were 2 minutes and 30 minutes, respectively.

After the sedimentation time of 1h, approximately 500 mL of the supernatant liquid was collected and the following parameters were analyzed: COD, chlorides, ammonia, and alkalinity by titulometry; nitrite, nitrate, and phosphate by spectrophotometry in UV-160ª-Shimadzu spectrophotometer; pH with digital pH measuring tool MB10-Mars Scientific; turbidity by nephelometry (NTU); total solids, volatile and fixed by gravimetric method; all according to APHA, AWWA, WEF (Standard…, 1992).

It is emphasized that the adequacy of the pH of the crude effluent to the corresponding of the desired treatment was performed with sulfuric acid solution ($H_2SO_4$) 0.02 N or sodium hydroxide (NaOH) 0.02 N, when necessary, being determined with the aid of pH meter.

## RESULTS AND DISCUSSION

Tables 1, 2, 3 and 4 show the statistical analysis and Tukey's test of the treatments and the dosages of chitosan used for the parameters analyzed, calculated from the analyses of the collected samples.

According to the Copam/CERH-MG Joint Regulatory Resolution No. 01 of May 5, 2008, effluents from any polluting source can only be released directly to the receiving body after proper treatment and if they meet the conditions, standards and requirements laid. The parameters COD, BOD and pH of the raw effluent are inadequate according to this Regulatory Resolution release pattern, so some type of treatment should be adopted before releasing it into the water body. The alkalinity, turbidity, solids, and phosphate parameters do not present maximum values established by current legislation. Thus, there is no pattern for comparing what can be released and the collected raw effluent.

Table 1. Mean values of COD, TS, TVS, TFS and turbidity parameters evaluated in each treatment

| Treatment | pH | COD (mg $L^{-1}$) | TS (mg $L^{-1}$) | TVS (mg $L^{-1}$) | TFS (mg $L^{-1}$) | Turbidity (NTU) |
|---|---|---|---|---|---|---|
| 1 | 5,5 | 4125.00a | 468.57a | 306.86a | 161.71a | 64.14a |
| 2 | 6,0 | 3825.00a | 851.43b | 611.86b | 239.57a | 218.77b |
| 3 | 6,5 | 4773.21a | 996.57b | 495.57b | 627.00b | 137.30ab |

Means followed by the same letter do not differ from each other by the Tukey's test (P>0.05).

Table 2. Mean values of alkalinity, chlorides, ammonia, nitrite, nitrate and phosphate parameters evaluated in each treatment

| Treatment | pH | Alkalinity (mg $L^{-1}$) | Chlorides (mg $L^{-1}$) | Ammonia (mg $L^{-1}$) | Nitrite (mg $L^{-1}$) | Nitrate (mg $L^{-1}$) | Phosphate (mg $L^{-1}$) |
|---|---|---|---|---|---|---|---|
| 1 | 5,5 | 668.57b | 22.29a | 66.8b | 0.0065a | 12.85a | 4.31a |
| 2 | 6,0 | 460.71a | 23.14a | 27.2a | 0.0137a | 25.84a | 4.79a |
| 3 | 6,5 | 630.00ab | 37.00b | 84.4b | 0.0098a | 18.73ab | 4.77a |

Means followed by the same letter do not differ from each other by the Tukey test (P>0.05).

Table 3. Mean values of COD, TS, TVS, TFS and turbidity parameters in each chitosan dosage

| Stitches | Chitosan dosage | COD (mg $L^{-1}$) | TS (mg $L^{-1}$) | TVS (mg $L^{-1}$) | TFS (mg $L^{-1}$) | Turbidity (NTU) |
|---|---|---|---|---|---|---|
| P1 | Raw Effluent | 9687.5b | 1317b | 681a | 717b | 323.75b |
| P2 | 0.25 g $L^{-1}$ | 4025.0ab | 710a | 441a | 265a | 131.00ab |
| P3 | 0.50 g $L^{-1}$ | 2475.0a | 705a | 453a | 241a | 149.00ab |
| P4 | 0.75 g $L^{-1}$ | 3325.0a | 749a | 514a | 283a | 105.00a |
| P5 | 1.00 g $L^{-1}$ | 3712.5a | 657a | 412a | 294ab | 93.00a |
| P6 | 1.25 g $L^{-1}$ | 3512.5a | 620a | 411a | 275a | 89.00a |
| P7 | 1.50 g $L^{-1}$ | 2950.0a | 647a | 388a | 324ab | 90.00a |

Means followed by the same letter do not differ from each other by the Tukey's test (P>0.05).

Table 4. Mean values of alkalinity, chlorides, ammonia, nitrite, nitrate and phosphate parameters in each chitosan dosage

| Stitches | Chitosan dosage | Alkalinity (mg $L^{-1}$) | Chlorides (mg $L^{-1}$) | Ammonia (mg $L^{-1}$) | Nitrite (mg $L^{-1}$) | Nitrate (mg $L^{-1}$) | Phosphate (mg $L^{-1}$) |
|---|---|---|---|---|---|---|---|
| P1 | Rough Effluent | 705a | 32a | 75a | 0.02a | 33a | 6a |
| P2 | 0.25 mg $L^{-1}$ | 580a | 25a | 51a | 0.02a | 18a | 5a |
| P3 | 0.50 mg $L^{-1}$ | 580a | 26a | 62a | 0.01a | 14a | 5a |
| P4 | 0.75 mg $L^{-1}$ | 555a | 28a | 60a | 0.01a | 18a | 4a |
| P5 | 1.00 mg $L^{-1}$ | 565a | 27a | 62a | 0.01a | 14a | 4a |
| P6 | 1.25 mg $L^{-1}$ | 555a | 28a | 58a | 0.01a | 17a | 4a |
| P7 | 1.50 mg $L^{-1}$ | 565a | 27a | 49a | 0.01a | 21a | 4a |

Means followed by the same letter do not differ from each other by the Tukey's test (P>0.05).

The best efficiency of removal of parameters using chitosan was found in treatment 1, followed by treatment 2. This can be explained, because according to Huang and Chen (1996), at a pH below 8.7 the amino groups of the chitosan polymer chain are completely protonated favoring the agglutination of particles. Muzzarelli and Rocchetti (1986) state that pH's below 6.5 aid in the process of coagulation by chitosan, as it presents a good solubility in acidic media.

The results of the COD parameter for the treatments did not show significant difference. The dosages of 0.5 to 1.5 g $L^{-1}$ of chitosan did not present statistical difference among themselves, but presented difference between P2 and P1, which reinforces the use of coagulant to reduce the values of this parameter.

The value of COD for the release of effluents allowed by joint COPAM/CERH-MG (Minas Gerais, 2008) is 180 mg $L^{-1}$. The results of the raw effluent and effluent after the use of the coagulant are outside the allowed limits. However, using 0.5 to 1.5g $L^{-1}$ chitosan, the removal of COD was around 67.6%, which shows the potential efficiency in coagulant use. Even though the product did not meet the discharge standard, it could be used as a primary treatment, followed by another treatment that would meet the COD standards.

Regarding the efficiency already recorded of the use of this natural coagulant, Vive and Yamaguchi (2011) using dosages of 0.4; 0.8 and 1.2g $L^{-1}$ for controlled landfill slurry treatment showed that the efficiency of COD removal at pH 3 was approximately 7; 2 and 1%. For pH 4 the efficiencies were approximately 41; 27 and 21%. At pH 5 the efficiencies were approximately 9; 18 and 10%, respectively, including that for better efficiency of COD removal (41%) a concentration of 0.4g $L^{-1}$ for pH 4 should be used. Using a dosage similar to that of the author above, 0.5g $L^{-1}$, and a pH between 5.5 and 6, 77.5% of COD removal was obtained.

Pacheco-Aguilar *et al*. (2009) performed tests using chitosan, after centrifugation of the sample, to treat glue water from the sardine industry. The authors obtained 31.2% efficiency of COD removal using 1.0g $L^{-1}$ chitosan, and the pH used (4.0, 5.0, 6.0 and 7.0) did not show significantly different results. Hassan *et al.* (2009) used 30mg $L^{-1}$ and pH 4 to treat effluent from the textile industry and obtained approximately 72% of COD removal. Belli *et al.* (2012), using 1g $day^{-1}$ chitosan as an auxiliary measure in the removal of organic matter and nutrients, in the membrane bioreactor, used to treat sanitary sewage, obtained 97% of COD removal. All these authors corroborate the idea that the use of chitosan to remove COD is effective for the effluents mentioned and for the effluent of the fish processing industry.

For TS, both the use of the different chitosan dosages and the treatments gave significantly different results, where T2 and T3 were statistically equal, differing from T1, which obtained better results. Chitosan dosages did not present statistical differences between them, but showed differences in relation to P1, which states the possible use of chitosan for removal of total solids.

Using the lowest chitosan dosage tested, 0.25 g $L^{-1}$, the results were statistically equal and test removal efficiency of TS found was 46.06%, a result higher than that found by Pacheco-Aguilar *et al.* (2009) who performed tests using chitosan, after centrifugation of the sample, in glue water of the sardine industry. The authors obtained 33.88% of TS removal after centrifugation and with the use of 1.0g $L^{-1}$ chitosan, this parameter was reduced by more than 25% at pH 4. Pawlowsky and Prado (2001) studied the use of chitosan treating water from the cassava starch refining process, obtaining a removal efficiency of 65.68% of TS using 3720.2 mg $L^{-1}$ from coagulant. These studies confirm the fact that it is possible to use chitosan, with good values of removal of TS.

For TVS, the treatments were significant, where T1 presented better results than T2 and T3, statistically equal. The dosages did not present significant difference in relation to P1, showing the non-interference in the removal of TVS. Although the present study did not show the efficiency of use of chitosan for removal of TVS, Pawlowsky and Prado (2001) studied its use (2688.8 mg $L^{-1}$ chitosan) to treat water from the cassava starch refining process, obtaining an efficiency of 65.9% removal. Compared to other treatment, Pereira *et al*. (2011) tested the efficiency of an anaerobic reactor system in the treatment of liquid swine effluents, obtaining 45% efficiency of TVS removal. Although the use of chitosan presented removal between 35.3% using 0.25 g $L^{-1}$ and 42.98% using 1.5 g $L^{-1}$ the results are lower than those found by the cited authors. Fact that does not exclude its use to remove TVS from the effluent processing of fish.

For TFS, both the use of the different chitosan dosages and the treatments gave significantly different results, where T1 and T2 were statistically equal, presenting better results than T3. P2, P3, P4 and P6 were statistically equal, differing from P1, which presented values statistically equal to P5 and P7.

Comparing with another type of treatment system, Pereira *et al*. (2011) testing the efficiency of an anaerobic reactor system in the treatment of liquid swine effluents, obtained 51% efficiency of TFS removal. Results lower than that found in this study, where with the use of 0.5 g $L^{-1}$ chitosan, was obtained a removal of 66.36%.

For alkalinity the treatments were significant, where T1 and T2 are significantly distinct, but each being equal to T3. Chitosan dosages did not present significant differences, including in relation to P1, showing that they do not interfere with alkalinity. Studies with chitosan use for this parameter did not were found, but Silva *et al*. (2007) using *Oleiferous moringa*, a natural coagulant, also did not find any significant change in the alkalinity of the medium, which in this case was used as a treatment of effluents from anaerobic reactors in the dosage of 200 mg $L^{-1}$ of this coagulant.

For turbidity, treatments T1 and T2 are significantly different, but each being equal to T3. Chitosan dosages showed significant differences in the results. The dosages of 0.75 to 1.5 g $L^{-1}$ were statistically equal, differing from the dosages of 0.25 and 0.5g $L^{-1}$ and P1, which are statistically equal. As expected, the use of chitosan showed satisfactory results for turbidity removal, with a dose of 0.75g $L^{-1}$ and a removal of 67.5% indicating good efficiency.

Kawamura (1991) evaluated the use of chitosan as coagulant with different surface waters with turbidity between 10 and 15 NTU and showed that with dosages lower than 5 mg $L^{-1}$ the residual turbidity was less than 1 NTU. Patel and Vashi (2012) used chitosan in a textile effluent with a dye concentration of 200mg $L^{-1}$, achieving a turbidity removal of 67%, witha dosage of 25mg $L^{-1}$ and pH 4. For the same type of effluent, Hassan *et al*. (2009) used 30mg $L^{-1}$ at pH 4, obtaining approximately 95% turbidity removal. These authors attest to the possible use of chitosan in the primary treatment by coagulation.

For T1 and T2 chlorides were statistically equal, presenting better results than T3. Chitosan dosages did not present significant difference, including P1, showing its non-interference in chloride removal. No studies were found using chitosan to remove this parameter, but Mounteer *et al*. (2007) treating effluents from the textile and kraft cellulose industries by combining aerobic biological treatment with advanced oxidative processes to intensify the removal of organic matter, achieved a 36% reduction in chloride content.

For ammonia, the treatments were significant, being T1 and T3 statistically equal, differing from T2, which presented better results. Chitosan dosages did not show significant differences, including in relation to P1, showing that the dosages do not interfere with ammonia removal. Although it did not find efficiency of the use of chitosan for ammonia removal, Belli *et al.* (2012) using chitosan, in the removal of organic matter and nutrients, in a membrane bioreactor, for sanitary sewage, with 1.0g $day^{-1}$ obtained 99.5% ammonia removal.

For nitrite the treatments did not differ in relation to P1, showing that this does not interfere with nitrite removal. For nitrate, treatments T1 and T2 are significantly different, but each in turn equal to T3. Chitosan dosages showed no significant differences, including in relation to P1, showing that they did not interfere with nitrate removal. For removal of ammonia, nitrite and nitrate the most recommending would be the use of a biological treatment.

For phosphate, the treatments showed equal results. Chitosan dosages also did not have significant differences, including in relation to P1, showing that they do not interfere in phosphate removal.

## CONCLUSION

The use of chitosan as a coagulant for effluents from the fish industry is satisfactory for use as a primary treatment. In the work in question, the pH values 5.5 and 6.0 showed the best responses to the use of chitosan. At dosages between 0.25 g L-1 and 1.5 g L-1, the use of chitosan was not active in the parameters TVS, alkalinity, chlorides, ammonia, nitrite, nitrate, and phosphate. However, for the parameters COD, TS, TFS and turbidity, chitosan action was identified at the same dosages. Thus, the best chitosan dosage indicated for the treatment of fish effluent was 0.25 g L-1 with optimized activity at a pH of 5.5.

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