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A comparative study of the adsorption of azo dyes in mixed adsorbents composed of *Aspergillus niger* and *Citrus sinensis* chemically modified: Influence of pH

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Abstract. The economic growth of the textile sector in Brazil has contributed to the generation of new jobs, qualification of the workforce and better living conditions for the population. However, due to the high activity of the sector, large volumes of toxic effluents have been generated which, if improperly disposed of, cause serious environmental damage. Therefore, alternatives in the treatment of effluents are interesting to increase the availability of this resource. A technique that has stood out, for being efficient, easy to operate and economical, is adsorption, a passive capture process where pollutants are deposited on the surface of materials. Thus, the work aimed to carry out a comparative study of the influence of pH in the removal of the dyes Remazol Black (RB), Remazol Red (RR) and Remazol Yellow (RGY) using different mixed adsorbents constituted by the fungus *Aspergillus niger* grown in orange peel in two different culture media. Six types of adsorbents (three for each culture medium) were produced and were treated with acid, base and without treatment. These were subjected to adsorption tests at different

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pHs (2, 7 and 9), using a solution (25 mg·L^{-1}) volume of 100 mL, 0.5 g of biomass, rotation 150 rpm, 30 °C. The results showed that the adsorbents produced were able to adsorb the dyes RB, RGY, RR at different pHs. The best adsorption condition was obtained at pH 2 and the adsorbents were treated with acid, showing a promising alternative for the treatment of textile effluents.

Keywords: Effluent treatment; Water quality; Textile dyes.

Resumo. ***Estudo comparativo da adsorção de corantes azo em adsorventes mistos compostos por* Aspergillus niger *e* Citrus sinensis *quimicamente modificados: influência do pH*.** O crescimento econômico do setor têxtil no Brasil tem contribuído para a geração de novos empregos, qualificação da mão de obra e melhores condições de vida para a população. Entretanto, devido à alta atividade do setor, tem se gerado grandes volumes de efluentes tóxicos que se descartados de forma inadequada causam sérios danos ambientais. Sendo assim, alternativas no tratamento de efluentes são interessantes para aumentar a disponibilidade deste recurso. Uma técnica que vem se destacando, por ser eficiente, de fácil operação e econômica é a adsorção, um processo de captação passiva onde os poluentes depositam-se na superfície de materiais. Assim, o trabalho teve como objeto realizar um estudo comparativo da influência do pH na remoção dos corantes Remazol Black (RB), Remazol Red (RR) e Remazol Yellow (RGY) utilizando diferentes adsorventes mistos constituídos pelo fungo *Aspergillus niger* crescido em casca de laranja em dois diferentes meios de cultura. Foram produzidos seis tipos de adsorventes (três para cada meio de cultura) e foram submetidos a tratamento com ácido, base e sem tratamento. Estes foram submetidos a ensaios de adsorção em diferentes pHs (2, 7 e 9), utilizaram-se um volume de solução (25 mg·L^{-1}) de 100 mL, 0,5 g de biomassa, agitação 150 rpm, 30 °C. Os resultados mostraram que os adsorventes produzidos foram capazes de adsorver os corantes RB, RGY, RR em diferentes pHs. A melhor condição de adsorção foi obtida em pH 2 e os adsorventes tratados com ácido. Sendo assim estes adsorventes tem se mostrando uma alternativa promissora para o tratamento de efluentes têxteis.

Palavras-chave: Tratamento de efluentes; Qualidade da água; Corantes têxteis.

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Introduction

The textile sector is one of the most complex and traditional sectors on the planet (Panigrahi et al., 2020). Its production chain begins with the production of fibers and filaments, including spinning, weaving, knitting, finishing and making (Basak et al., 2020). Its industries, or at least part of them, are distributed all over the world, from countries with less economic development, as well as more developed ones (Basak et al., 2020; Srivastava et al., 2020; Silva et al., 2021).

Brazil is one of the largest textile producers in the world (ABIT, 2020). In 2019, an average of 8.9 billion pieces were produced in the country (clothing, accessories, bed, table and bath), the sector generated an average of 1.5 million direct employees and 8 million indirectly and had revenues of US$ 48.3 billion (ABIT, 2020). The textile hubs of the Brazil that deserve to be highlighted are Ceará, Agreste Pernambucano, Vale do Itajaí, in the State of Santa Catarina, and the Americana hub, in São Paulo (FEBRATEX, 2020a).

The center located in Agreste Pernambucano, formed mainly by the cities of Santa Cruz do Capibaribe, Toritama and Caruaru, is the second largest producer of clothing in the country, according to the National Service for Industrial Learning (SENAI), second only to São Paulo (FEBRATEX, 2020b; Esteves, 2020). The region receives customers from all over the country, predominantly from the North and Northeast States (Esteves, 2020). In recent years, the demand for buyers from the Midwest and, even more, the Southeast has grown, like Minas Gerais and Espírito Santo (Esteves, 2020; FEBRATEX, 2020b). The high shopping season occurs in June and July, and from November to December (Esteves, 2020).

The beauty of the textile sector is in the pieces produced, these have strong and vibrant colors which draw the attention of the consuming public (Srivastava et al., 2020). Among the different dyes used in dyeing are the azo dyes (-N=N-), they are widely used by the textile industries because they are easy to produce, low cost and present a wide variety of colors, in addition to being soluble in water (Nazir et al., 2020). However, during the dyeing process large volumes of water are used, resulting in a colored effluent that, when improperly disposed of, causes serious problems to both human health and the aquatic ecosystem (Song et al., 2020).

The treatment of effluents containing azo dyes has become a worldwide challenge, since many physical and chemical methods do not completely remove this type of dye, besides being operationally expensive methods, and with the generation of quite toxic by-products (Panigrahi et al., 2020). So, adsorption has become an effective alternative for the removal of these pollutants in water, since it is a simple technique, easy to operate and low cost (Nazir et al., 2020). Different adsorbent materials have been studied, however, the ones that have gained prominence due to the abundance and also the low cost are the natural adsorbents (Khalaf, 2008; Saratale et al., 2013; Huang et al., 2016). Different adsorbent materials can be used to remove dyes, macadamia seed peels (Felista et al., 2020), chitosan (Ammadurai et al., 2007), *Aspergillus niger*/elephant grass (Cruz-Filho et al., 2016), and activated carbon derived from coconut shell (Furlan et al., 2010).

Therefore, this study aimed to evaluate the influence of pH in the removal of the dyes Remazol Black, Remazol Red and Remazol Yellow, using as mixed adsorbent the mixed biomass composed by the fungus *Aspergillus niger* grown in orange peel, in order to obtain higher percentages removal and decrease costs with the treatment of textile effluents.

Materials e methods

Adsorbate

The dyes used in this work were: Remazol Black B (RB) (Figure 1A), Remazol Golden Yellow RNL (RGY) (Figure 1B) and Remazol Red (RR) (Figure 1C) at maximum wavelengths 597, 410 and 518 nm, respectively (Aksu et al., 2005).

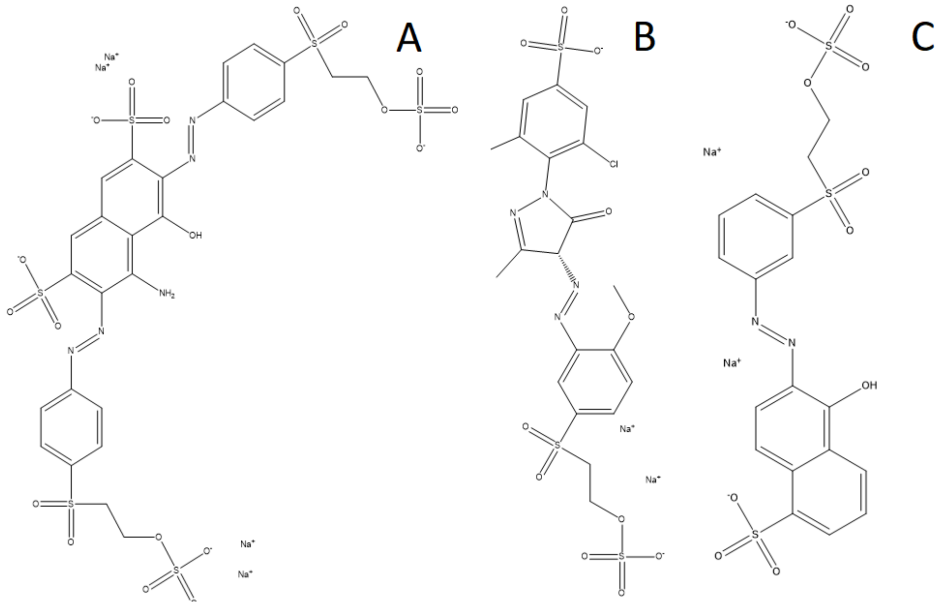

Figure 1. Chemical structure of textile dyes. Remazol Black B (A), Remazol Golden Yellow RNL (B) and Remazol Red (C)

For the construction of the analytical curves, 100 mL of stock solutions of the dyes were prepared in a concentration of 200 mg·L^{-1}. From this concentration, different dilutions were performed to reach concentrations from 0 to 105 mg L^{-1} and, in this way, build the calibration curve. After being prepared, an aliquot was removed from each solution for later absorbance reading, in a spectrophotometer (Hewlett-Packard, model 8453), at the respective wavelengths. Distilled water was used as the white of the equipment. The data obtained were used to construct the graphs of the calibration curves.

Study of the stability of dye solutions at different pH

The evaluation of the degradation of the dye (in relation to time) was carried out according to Zanon (2006) with modifications. This study was carried out using different solutions of the dyes (RB, RGY and RR) in a concentration of 25 mg·L^{-1}. For this experiment, 100 mL of dye solution were added to a 250 mL Erlenmeyer flask at 30 °C with agitation of 150 rpm at different pHs (2, 7 and 9). Every 20 min for 240 min, 3.0 mL aliquots were analyzed on a spectrophotometer.

Production of mixed adsorbent

The orange peels used in this work were obtained from snack bars located at 8° 02' 37.5" S and 34° 56' 57.3" W, Recife, Pernambuco State, Brazil. They were crushed in a blender, then they were cut, washed with distilled water and dried at 75 °C for 72 h. After drying, they were crushed and sieved again in Tyler sieves (1.43 mm). The microorganism used was the filamentous fungus *Aspergillus niger* (ATCC 1015) from the collection of microorganisms from the Industrial Microbiology Laboratory, of the Department of Chemical Engineering (DEQ), of the Federal University of Pernambuco (UFPE). The fungal mass was grown in Czapec-Dox medium for approximately 7 days in an oven at a temperature of 30 °C.

The methodology proposed by Cruz et al. (2016) was used to prepare the mixed adsorbent with few modifications. A volume of 5 mL of spore suspension (10^7 spores mL^{-1}) was inoculated in 250 mL in two different culture mediums, one with sucrose (MCS) consisting of $NaNO_3$ (3.0 g), KH_2PO_4 (1, 0 g), $MgSO_4.7H_2O$ (0.5 g), $FeSO_4$ (0.01 g), sucrose (3.3 g) and orange peels (6.7 g). The second without sucrose (MSS) composed of $NaNO_3$ (3.0 g), KH_2PO_4 (1.0 g), $MgSO_4.7H_2O$ (0.5 g), $FeSO_4$ (0.01 g) and orange peels (10 g). All experiments were carried out under aseptic conditions. After inoculation, the tests were kept under agitation at 200 rpm at 30 °C for 72 h. At the end of the cultivation, the material produced was autoclaved at 121 °C for 30 min, filtered and then washed with sterile water and dried in an oven at 75 °C.

The material produced by each of the MCS and MSS culture medium was divided into three aliquots and classified according to the chemical treatment submitted, thus having: i) samples without treatment; ii) treated with hydrochloric acid (HCl) 1.0 $mol{\cdot}L^{-1}$ for 1h and iii) treated with sodium hydroxide (NaOH) 1.0 $mol{\cdot}L^{-1}$ also for 1 h (Chaves et al., 2008; Cruz et al., 2016). After treatment, the samples were washed with plenty of distilled water to remove the treatment solution and taken to the oven at 75 °C for drying. Thus, six different adsorbents were produced: A1 (without treatment), A2 (acid) and A3 (basic) from the MCS medium and B1 (without treatment), B2 (acid) and B3 (basic) from the MSS medium.

Influence of pH in the removal process

To evaluate the influence of pH in the adsorption process, a methodology proposed by Cruz et al. (2016), Nascimento et al. (2017) and Molavi et al. (2018). For this, solutions of the dyes (RB, RGY and RR) were prepared at pH = 2 (acidic), pH = 7 (neutral) and pH = 9 (basic). The pH values were adjusted using solutions of sulfuric acid and 3M sodium hydroxide. The adsorption experiments were carried out in batches in Erlenmeyers flasks, containing 100 mL of the solution (25 $mg{\cdot}L^{-1}$) of each dye, 0.5 g of each mixed adsorbent, with fixed temperature and agitation of 30 °C and 150 rpm, respectively, for a period of 12 h. At the end of this test, an aliquot of the supernatant was removed and the final concentration of the dye present in the solution was analyzed in a UV-VIS spectrophotometer (Hewlett-Packard, model 8453) at their respective wavelengths. The tests were performed in triplicate and the percentage of dye removal (% R) was determined by Equation 1.

$$R(\%) = \left(\frac{C_0 - C_f}{C_0}\right) * 100 \tag{1}$$

Where C_0 and C_f are the initial and final concentrations of the dye in the liquid phase (mg/L) respectively.

Results and discussion

The calibration curves of the UV-VIS spectrophotometer for the dyes RB, RGY and RR at the wavelengths of 597, 410 and 518 nm are shown in Figure 2A, respectively. The results showed good linearity for the dyes under study. The results obtained, as shown in Figure 2B, 2C and 2D, demonstrate a small variation in the recorded absorbance, with no significant change in the dye structure. This is because the azo compounds are found in the form of hydrazo tautomer, its most stable form (Zanon, 2006; Nazir et al., 2020).

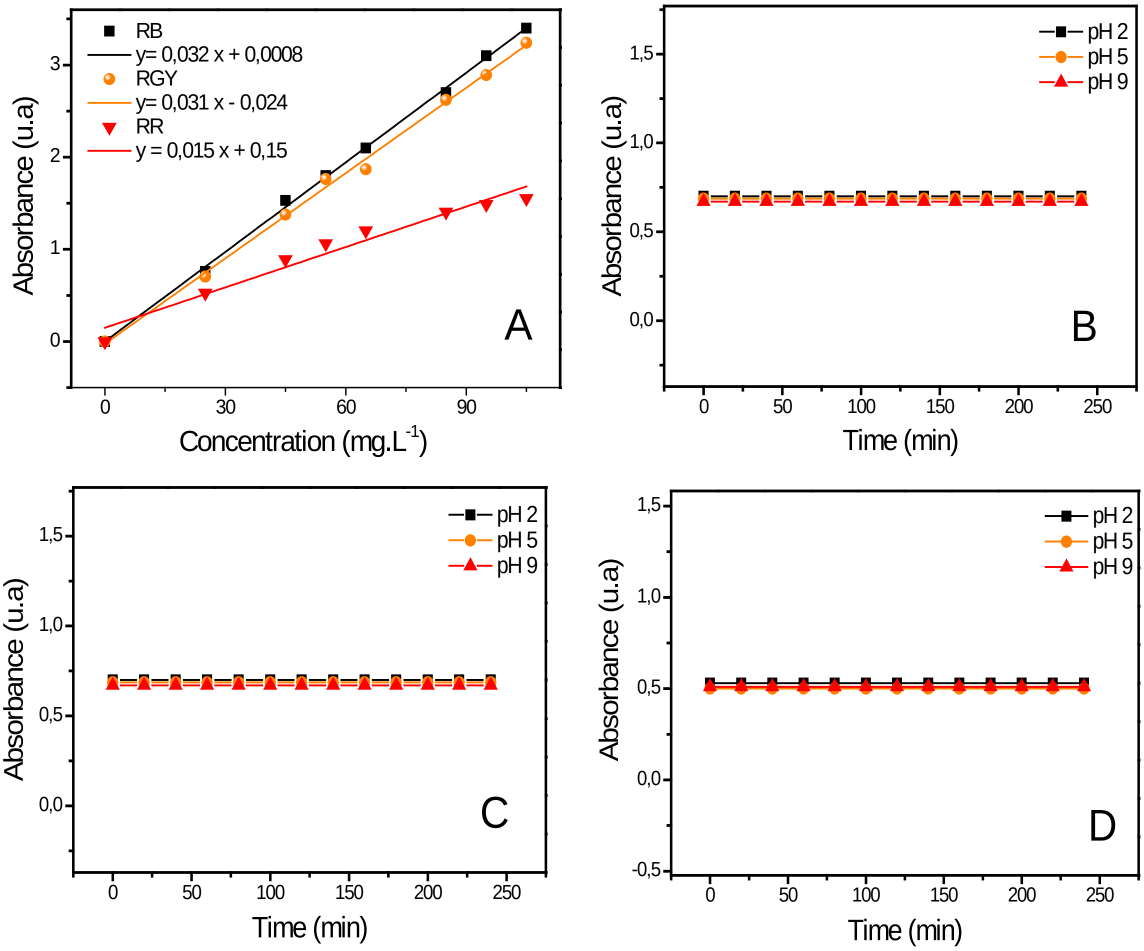

Figure 2. Calibration curve for the dye RB, RGY and RR (A), stability curves for the dye RB (B), (C) and (D).

The pH has a great influence on the dye adsorption process. Thus, a study was carried out to identify the best pH for carrying out the adsorption (Mahmoodi et al., 2011). The Figure 3 shows the percentage of removal of the adsorbents against the dyes RB, RGY and RR in relation to pH.

The results showed that all adsorbents produced showed higher percentages of removal at pH 2. For the RB dye (Figures 3A and 3B) the acid-treated adsorbent showed higher percentages of removal at all studied pHs. For Remazol Golden Yellow RNL (Figure 3C and 3D) the adsorbents showed a percentage of removal <50%, and the base-treated adsorbent removed even with less efficiency at all pHs. For Remazol Red (Figures 3E and 3F) the base-treated adsorbents also promoted adsorption in a greater pH range. Therefore, the best process condition for this study was pH 2 and acid-treated adsorbents. In this case, as the adsorbents produced in medium containing or not sucrose showed a significant difference in the adsorption process. Regarding the production cost of the adsorbent, the adsorbent produced in a medium without sucrose was chosen.

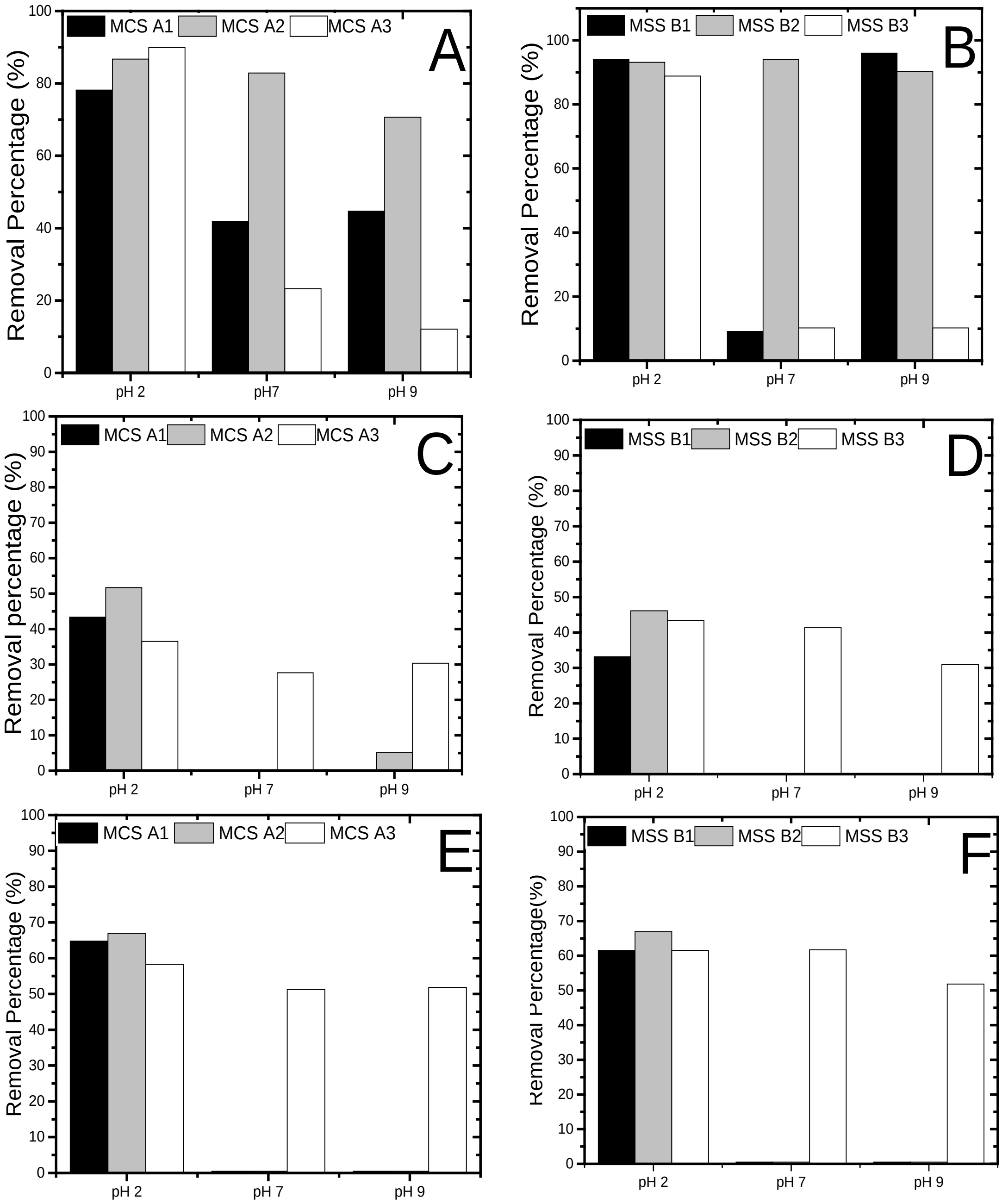

Figure 3. Comparison of the percentage of removal of adsorbents A1 (without treatment), A2 (acid) and A3 (basic) from the MCS medium. For SSS medium, adsorbents B1 (without treatment), B2 (acid) and B3 (basic) at different pH values: for Remazol Black B (A, B) Remazol Golden Yellow RNL (C, D) Remazol Red (E, F)

The adsorption mechanism between pH 1-4 is mainly due to electrostatic interactions. For this pH range, some of the groups of the adsorbent surface will be protonated and the adsorbent-dye interaction may occur via sulfonic groups of the dye (Mahmoodi et al., 2011). The Figure 4 represents the simplified electrostatic mechanism for this three-step interaction. In the first one, it can be seen that the hydroxyl groups of the adsorbent are protonated by the hydronic ions and dye molecules are dissociated by water, resulting in the interaction between opposite charges of the adsorbent and with the dye.

$$\text{Adsorvent}-OH + \overset{+}{O}H_3 \longrightarrow \text{Adsorvent}-\overset{+}{O}H_2 \quad (1)$$

$$\text{Dye} + H_2O \longrightarrow \text{Dye}-SO_3^- \quad (2)$$

$$\text{Adsorvent}-\overset{+}{O}H_2 + {}^{-}O_3S-\text{Dye} \longrightarrow \text{Adsorvent}-\overset{+}{O}H_2\text{-------}{}^{-}O_3S-\text{Dye} \quad (3)$$

Figure 4. Adsorption mechanism simplified in three stages showing the interaction via dye adsorption charges.

Other authors have also found similar behavior for the removal of RB, RGY and RR dyes. In studies with the RB dye. Cruz-Filho et al. (2016) also using mixed adsorbent composed of elephant grass and *Aspergillus niger* obtained removal around 96% in acidic conditions pH 2. Tunç et al. (2009) removing the same dye using cotton waste, stem and bark verified greater removals at pH 1.0. Ziapour et al. (2016) using sugarcane bagasse they verified that at pH 2.5 there is a removal around 55%. Hamzeh et al. (2011) using lignocellulosic residue from canola stems obtained removal around 80% at pH 2.5. Ucar, (2014), using pine needles obtained 91.51% removal at pH 3.0. Sukarta, (2020) obtained greater removal in pH 2 using cream of pin as adsorbent.

Regarding the RGY dye, satisfactory removals at acid pH were also obtained. Nascimento et al. (2014) using peanut peel and orange peel found greater removals at pH 2, with peanut peels being more efficient at removal. This same behavior was found by Nascimento et al. (2016), using banana peel and green coconut mesocarp the authors found that the green coconut mesocarp was more efficient in removal when compared to banana peels. Nugroho et al. (2008) evaluating a mixed adsorbent fungal biomass and activated carbon found better removal results at pH 1 (<95%) when compared to activated carbon at the same pH removal around 65%. This difference may be associated with the groups present in the fungal biomass.

In adsorption tests with RR dye, promising adsorbents were also found in the removal of the dye. Costa et al. (2019) obtained 90% removal at pH 2 using rice husk ash as an adsorbent. Chaudhuri et al. (2011), coconut activated carbon was used as an adsorbent removed around 80% of the dye over a wide pH range (1-10). Dey et al. (2016) found higher removal values at pH 3 with chitosan as an adsorbent. Ara et al. (2013) obtained 95% removal percentage at pH 2 using sawdust.

In addition to the adsorption study for the dyes under study in isolation, the literature presents comparative studies. Aksu et al. (2005) using seaweed *Chlorella vulgaris* as an adsorbent, he verified that the algae biomass removed the RB dye more efficiently, then the RR and RGY dyes. Thus, the differences in the removal of dyes by the adsorption process are directly related to the conditions of the process and the pH is only one parameter. The type of adsorbent, the quantity, the temperature, agitation, structural complexity of the dyes must be taken into account in order to obtain the best process condition (Arulkumar et al. 2011; Cruz-Filho et al., 2016).

Conclusion

Through this study, it can be concluded that through chemical modifications of the adsorbent surface it is possible to obtain biomaterials with different adsorptive capacity. The adsorbent *Aspergillus niger*/orange peel produced in different medium was able to adsorb different azo dyes (RB, RGY, RR) at different pHs. The best process condition was obtained at pH 2 and acid-treated adsorbents. In addition, the mixed adsorbent culture medium (with or without sucrose) did not influence the ability to capture the dyes through the substrate. Thus, the adsorbents produced are promising alternatives for efficient materials for the treatment of textile effluents.

Conflicts of interest

The authors declare that have no conflicts of interest.

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