

A comparison of mango seed kernel powder, mango leaf powder and *Manilkara zapota* seed powder for decolorization of methylene blue dye and antimicrobial activity

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Abstract

The waste mango seed generated from mango pulp industry in India is a major problem in handling the waste and hence, conversion of mango seed kernel. Mango seeds were collected and processed for oil extraction. Decolorization of methylene blue was achieved by mango seed kernel powder, mango leaf powder and *Manilkara zapota* seed powder. Higher efficiency was attained in mango seed kernel powder when compared to mango leaf powder and *Manilkara zapota* seed powder. A 60 to 95 % of removal efficiency was achieved by varying concentration. Effect of pH, dye concentration, adsorbent dosage and temperature were studied. Mango seed kernel powder is a better option that can be used as an adsorbent for the removal of methylene blue and basic red dye from its aqueous solutions.

Key words

Mango kernel, Mango leaf powder, *Manilkara zapota*

Introduction

India is the second largest producer of fruits and vegetables in the world. India is the world's largest producer of mangoes. India is the birth place of mangoes. Mango is known as the 'King of fruit' throughout the world. From the foothills of the Himalayas to the shores of the Indian Ocean, Indians have cultivated and nurtured mangoes for many centuries. Thus, mango has become the "National fruit of India" which is indigenous to the Indian Sub Continent. It belongs to the genus *Mangifera*, consisting of numerous species and also belongs to the flowering plant family Anacardiaceae (Nzikou *et al.*, 2009). India produced nearly 12 million metric tonnes of mangoes in the year 2010 accounting for more than 51.1% of total production of the world. As of FAO's statistics, India was the top producer and exporter of mangoes in 2014. Besides the export of mangoes as a fresh fruit, the other main mango products that we export are mango pulp, mango pickles and chutneys. The processed

mango pulp has enhanced shelf life and has significant export potential. India exported 64.5 lakh metric tonnes in the form of processed mangoes in the year 2014 given by APEDA (Agricultural and Processed Food Products Export Development Authority, India).

Dyes are synthetic organic compounds that are embedded with various functional groups. They are widely used in textile, leather, paper and plastic industries. Dyes are of different classes such as acidic, basic, direct, disperse, reactive, sulfur and vat. Among these classes the most commonly used dyes are acidic and basic dyes. These dyes degrade to produce carcinogens that produce toxic effects such as headache, dizziness, precordial fever, hypertension, necrosis, nausea, vomiting, abdominal pain, anemia, bladder irritation and discoloration of urine (Arogba *et al.*, 1997). The Indian Textile Industry has an overwhelming presence in the economic life of the country in which dyes are extensively used. The textile industry accounts for around 80% of

consumption in India (Hunger *et al.*, 2003). In addition to textiles, dyes find application in other industries like plastic, paints, printing inks, food processing, paper and leather industries. One of the major problems concerning to these industries are wastewaters as colored effluent. Direct depositions of these effluents into sewage networks produce disturbances in biological treatment processes and also impart color which is visible to human eye and therefore highly objectionable on aesthetic grounds (Hoda Roushdy *et al.*, 2010). They also interfere with the transmission of light and upset the biological metabolism processes which cause destruction of aquatic communities present in the ecosystem. If this pollution continues, then the surface water quality goes to the level at which it cannot be used for any other purpose. Currently, the major methods used for treating dye containing textile waste waters are physical and chemical processes such as chemical precipitation, air stripping, oxidation, flocculation, sedimentation, ion exchange and adsorption. However, these processes are effective and economic only in the case where solute concentrations are relatively high. Among these, the liquid-phase adsorption has proved to be an effective and attractive technique for removing dyes, odors and oil from aqueous solutions.

Neem (*Azadirachta indica*) leaf powder has been used as an adsorbent to study the adsorption kinetics and thermodynamics of methylene blue. Neem leaf powder was found to remove 93% of dye. (Bhattacharyya *et al.*, 2005). Wheat shells used as adsorbent was found to remove 96% methylene blue justifying that wheat shells can be an alternative instead of more costly adsorbents used for dye removal in wastewater treatment processes (Yasemin Bulut *et al.*, 2006, Weng *et al.*, 2009) had reported the ability of an unconventional bio-adsorbent, pineapple leaf powder for the adsorption of methylene blue from aqueous solution. Activated carbon prepared from agricultural waste material such as male flowers of coconut trees and the waste jute fibers from jute industry have been found with as a better adsorbent for removal of reactive red dye from aqueous solutions (Senthilkumar *et al.*, 2006).

Mango seed is an abundant residue discarded by mango juice manufacturing industries and its amount is increasing due to the expansion of fruit production. An alternative for the use of this residue is necessary and hence an attempt is made to utilize it as a source of cellulose. Mango seeds are used as cellulose sources for producing cellulose acetate asymmetric membranes.

In light of the above, a study was carried out to ascertain the ability of mango seed kernel powder, mango leaf powder and *Manilkara zapota* seed powder in decolorizing the methylene blue.

Materials and Methods

Seed powder : About 50 kg of mango seeds (waste) was collected from the local juice manufacturing industry Galla Foods Pvt. Ltd. The seeds were washed and air dried to remove the hard seed coat manually from the processed waste. These dried seeds were finely powdered using domestic blender into powdery form and sieved using a sieve of size 425 μ and that sieved particles *i.e.*, Mango seed kernel powder was used the further experiments. Similarly, *Manilkara zapota* seed was powdered and also used for further study.

Leaf powder: Leaves were dried to 104°C for 24 hrs and powdered by using mortar and pestle and preserved for further use.

Sample preparation : A 0.2g of sample was taken into 50ml of 100 mg l⁻¹ dye solution and kept in orbital shaker at 150 rpm for 30 min, so that the sample gets fixed onto the adsorption sites of taken dye solution. After removing from the orbital shaker, solution was taken and filtered by using Whatman 4 filter paper. Methylene blue, a heterocyclic cationic compound with molecular formula C₁₆H₁₈N₃SCl, was obtained from Thomas Baker Chemicals Pvt. Ltd. and similarly for basic red dye. The stock solution was prepared by dissolving 1g in 1000ml of distilled water.

Adsorption studies

Effect of pH : The percentage of dye removal and effect of pH were analyzed over the pH range of 2 to 10. pH was adjusted by using 0.1N NaOH and 0.1N HCl solutions. A 50ml of dye solution of 100 mg l⁻¹ concentration was agitated with 0.5 g of mango seed kernel powder and mango leaf powder in orbital shaker at 150rpm for about 1hr to know the effect of pH. The concentration of the residual methylene blue dye solution and basic red dye solution were analyzed by using UV100 series double beam Spectrophotometer at 650nm for methylene blue at fixed time intervals of 10, 20, 30, 40, 50 and 60 min.

Effect of initial dye concentration : The effect of initial dye concentration was estimated for every 10 min. by contacting 0.2 g of mango seed kernel powder and mango leaf powder with 50 ml of dye solutions at different initial concentrations ranging from 20 to 100 mg l⁻¹ for 60 min. For every 10 min, spectrophotometer analysis of supernatant was estimated residual dye concentration.

Effect of adsorbent dosage : The effect of adsorbent dosage on the amount of dye removal was obtained by agitating 50 ml of dye solutions of initial dye concentration of 100 mg l⁻¹ with weighed amount of mango seed kernel powder and

mango leaf powder (ranging from 0.1 to 1.0 g) for about 60 min. Residual dye concentration of supernatant was analyzed for every 10 min by UV-Spectrophotometer.

Effect of temperature : The effect of temperature was studied at 303K, 313K and 323K to study the effect of mango seed kernel particles' on adsorption rate of methylene blue dye solutions. The effect of temperature was estimated by contacting 0.2g of mango seed kernel powder and mango leaf powder with 50 ml of dye solution of different dye concentrations such as 20, 40, 60, 80, and 100 mg l⁻¹. Supernatant was collected and UV-Spectrophotometer analysis was done to know the residual dye concentration.

Antimicrobial activity : The antibacterial and antifungal activities were evaluated with ethanolic extract of mango (*Mangifera indica*) kernels and mango leaf powder extract by Well diffusion method, against the most prevalent food borne

pathogens such as *Klebsiella pneumonia*, *Pencillium chrysogenum*. 1.3 g of HIMEDIA instant nutrient broth supplemented with nutrients was dissolved in 100 ml of distilled water. The prepared broth was sterilized and stored at 4°C until required. Bacterial and fungal strains were sub-cultured on to their respective growth medium and then incubated for 48 hrs at 25-30°C. From these plates, several colonies were transferred to 5 ml of sterile distilled water. The suspensions were mixed for 15 sec. to ensure homogeneity and then sterilized and supplemented to working suspension. 5g of nutrient agar (Muller Hinton Agar) medium available instantly was dissolved in 200ml of distilled water and autoclaved along with petri plates. Then the medium was taken from autoclave and kept in laminar air flow chamber and poured into sterilized plates for solidification. After solidification, the zone of inhibition was measured after incubation.

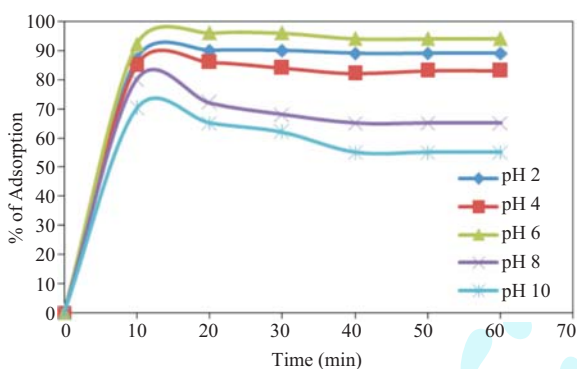


Fig. 1 : Effect of pH against time for methylene blue by mango seed kernel powder (Initial dye concentration-100 mg l⁻¹; adsorbent dose-0.5 g 50 ml⁻¹; equilibrium time-1hr)

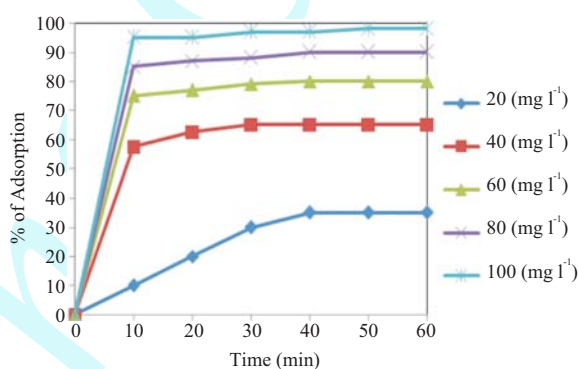


Fig. 2 : Percentage of adsorption with effect of Initial dye concentration for methylene blue by mango seed kernel powder (adsorbent dose-0.2 g 50 ml⁻¹; equilibrium time-1 hr)

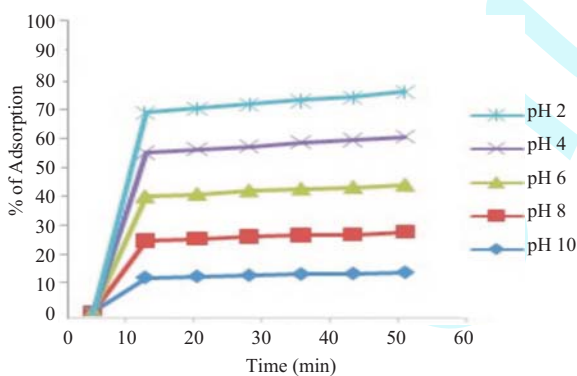


Fig. 1(a) : Effect of pH against time for methylene blue by *Manilkara zapota* seed Powder (Initial dye concentration-100 mg l⁻¹; adsorbent dose-0.5 g 50 ml⁻¹; equilibrium time-1hr).

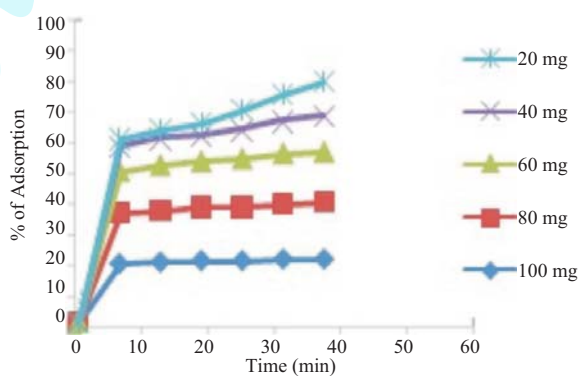


Fig. 2(a) : Percentage of adsorption with effect of Initial dye concentration for methylene blue by *Manilkara zapota* seed powder (adsorbent dose-0.2 g 50 ml⁻¹; equilibrium time-1hr)

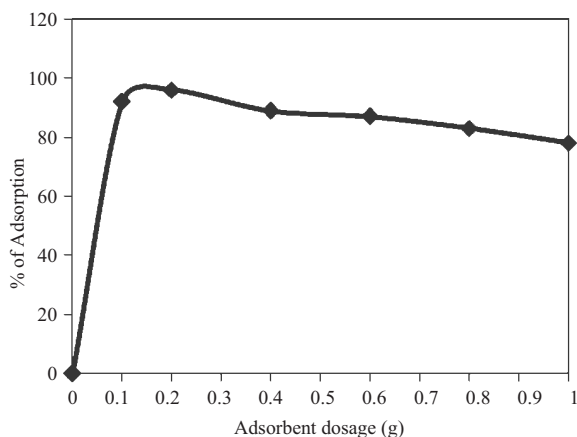


Fig. 3 : Percentage of adsorption with effect of adsorbent dosage for methylene blue by mango seed kernel powder (Initial dye concentration-100 mg l⁻¹; Equilibrium time-1hr)

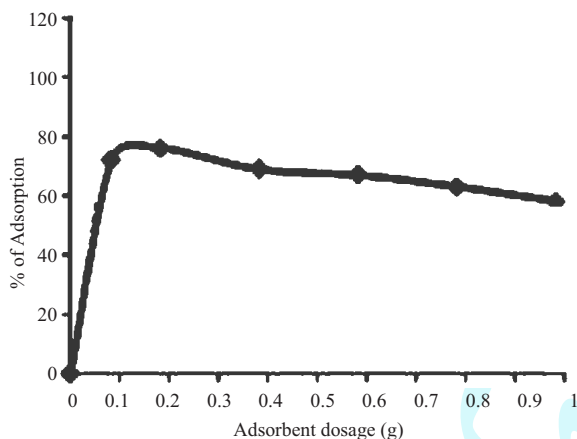


Fig. 3(a) : Percentage of adsorption with effect of adsorbent dosage for methylene blue by *Manilkara zapota* seed powder (Initial dye concentration-100 mg l⁻¹; equilibrium time 1hr)

Results and Discussion

Effect of pH : pH plays an important role in the adsorption of solutes from aqueous solutions. The adsorption capacity, chemistry of dye molecules and behavior of adsorbent were controlled by pH. The pH varied from 2 to 10. Fig. 1 and Fig. 1 (a) depicts the effect of pH on the percentage of adsorption of methylene blue by mango seed kernel powder. Maximum of 97% of adsorption was observed at pH 6 and minimum percentage of adsorption of 55% adsorption was seen at pH 10. The amount of dye adsorbed decreased from 22.5mg g⁻¹ to 17.5mg g⁻¹. Due to a significant electrostatic attraction between the cationic dye and negatively charged surface of the adsorbent. pH 6 with maximum adsorption was taken as optimum pH, as it did

not require any pretreatment. Hence further experiments were carried out at this pH. For mango leaf powder, the adsorption percentage was 68% and for *Manilkara zapota* seed powder, the adsorption percentage was 75% at pH 2 and 54% in pH 10, respectively.

Effect of initial dye concentration : The initial dye concentration had a pronounced effect on its removal from aqueous solutions. The effects of contact time on the adsorption of methylene blue and basic red dye at different initial dye concentrations (20 mg l⁻¹, 40 mg l⁻¹, 60 mg l⁻¹, 80 mg l⁻¹, 100 mg l⁻¹) onto the adsorbent mango seed kernel and *Manilkara zapota* seed powder and results are presented in (Fig. 2 and 2(a)). It was found that the removal of dye increased from 10% to 98% for methylene blue by mango seed kernel powder and 54% achieved for mango leaf powder. For *Manilkara zapota* seed powder, the removal of dye increased from 10-72% for methylene blue.

For methylene blue dye, the amount of dye adsorbed increased from 0.5 mg g⁻¹ to 24.5 mg g⁻¹ and for basic red dye adsorption increased from 7.5 mg g⁻¹ to 24 mg g⁻¹. The percentage of adsorption increased with initial dye concentration because, the initial dye concentration provided the driving force to overcome the resistance to the mass transfer of dye between the aqueous and the solid phase. The increase in initial dye concentration also enhanced the interaction between adsorbent and dye. Higher the methylene blue concentration, higher was the adsorption due to the driving force of concentration gradient that created more number of vacant sites. Apparently, this resulted in increased adsorption between the adsorbate in the solution and adsorbate in the adsorbent.

Effect of adsorbent dosage : The capacity of adsorbent is strongly affected by the amount of dosage. A series of experiments were performed to know the effect of adsorbent dosage by maintaining at pH 6 with optimum concentration of 100 mg l⁻¹ under equilibrium time of 1 hr by varying the amount of adsorbent dosage from 0.1g to 1.0g. Fig. 3 shows that the percentage of adsorption for methylene blue dye solution decreased from 84% to 74% with increase of adsorbent from 0.1 g to 1.0 g. At 0.2 g, highest adsorption of 97% was observed. Which might be due to fact that certain amount of dye could adsorb only fixed amount of adsorbent. The amount of dye adsorbed decreased from 23.5 mg g⁻¹ to 3.95 mg g⁻¹. As highest percentage of adsorption occurred at 0.2g when compared to other dosages it was taken as optimum dosage. The concentration gradient between adsorbent surface and solute concentration was the driving force for decrease in the amount of dye adsorbed with increasing adsorbent mass. For mango leaf powder, the efficiency was 62% and for *Manilkara zapota* seed powder higher adsorption was attained at 0.10g (Fig. 3a).

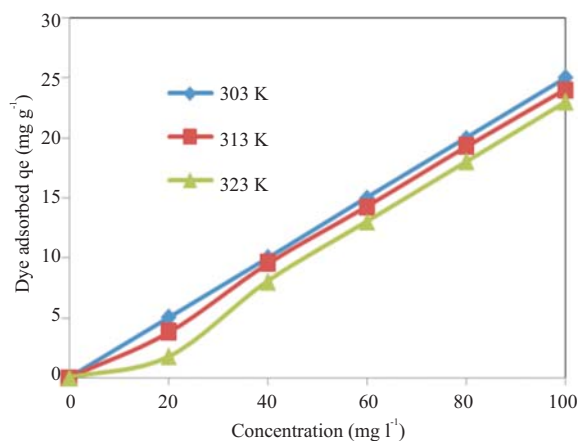


Fig. 4 : Effect of temperature against concentration for methylene blue by mango seed kernel powder (Initial dye concentration-100 mg l⁻¹; Adsorbent dosage-0.2 g 50 ml⁻¹; Equilibrium time-1hr)

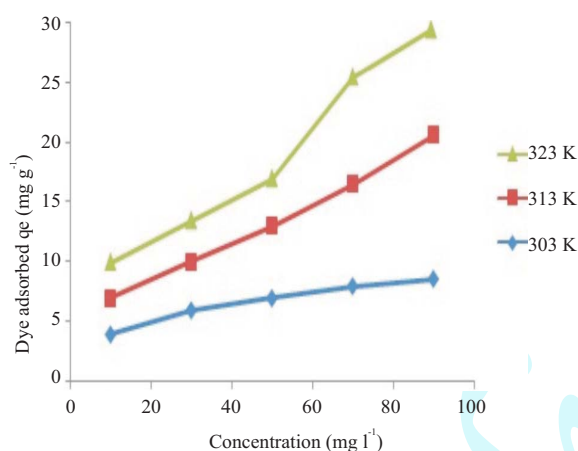


Fig. 4(a) : Effect of temperature against concentration for methylene blue by *Manilkara zapota* seed powder (Initial dye concentration-100 mg l⁻¹; Adsorbent dosage-0.2 g 50 ml⁻¹; Equilibrium time 1hr)

Effect of temperature : The effect of temperature on adsorption rate of methylene blue on to mango seed kernel particles was investigated at 303 K, 313 K and 323 K. The temperature has two important effects on the adsorption process. Increasing the temperature is known to increase the rate of diffusion of the adsorbate molecules across the external boundary layer and the internal pores of the adsorbent particle, owing to the decrease in the viscosity of the solution for highly concentrated suspensions (Mehmet Dogan *et al.*, 2009). Consequently, change in temperature would change the equilibrium capacity of the adsorbate.

Fig. 4 shows variation in dye adsorption for methylene blue by mango seed kernel powder dye solution with variation in concentration. This indicates that the



Fig. 5 : Antimicrobial activity of *Klebsiella pneumoniae*



Fig. 6 : Antimicrobial activity of *Penicillium chrysogenum*

amount of dye adsorbed increased from 5 mg g⁻¹ to 23mg g⁻¹ with increase in temperature, ensuring that the process was endothermic. As a matter of fact, the interaction between functional groups and mango seed kernel particle were favored at high temperatures, and for mango kernel it was 59%. For *Manilkara zapota* seed powder, the amount of dye adsorbed varied from 5 to 20 mg g⁻¹ (Fig. 4a).

Adsorption is known to be a promising technique, which has great importance due to the ease of operation and comparably low cost of application in the decoloration

process from contaminated water. Much attention has been focused on various naturally occurring adsorbents such as chitosan, zeolites, fly ash, coal, paper mill sludge, and various clay minerals (Rafatullah *et al.*, 2010 Unuabonah *et al.*, 2004).

An attempt to develop cheaper and effective adsorbents and many non-conventional low-cost adsorbents such as clay materials, zeolites, siliceous material, agricultural wastes and industrial waste products have also been suggested (Crini *et al.*, 2006, Rauf *et al.*, 2009).

Excellent ability and economic promise of adsorbents prepared from biomass exhibited high sorption properties from various sources. With the recent development on the use of low cost adsorbent, a wide range of current researches on non-conventional adsorbents like mango seed kernel powder (Kumar *et al.*, 2005; Bulut *et al.*, 2006), wheat shell (Bulu *et al.*, 2006), neem leaf powder (Bhattacharyya *et al.*, 2005), jute fibre carbon (Senthilkumar *et al.*, 2005), rice husk (Vadivela *et al.*, 2005), giant duckweed, date pit, wool fibre (sheep wool), cotton fibre (Waranusantigul *et al.*, 2003, Khan *et al.*, 2005) to enlighten researchers on the adsorption capacities of different biological material have been carried out.

The methylene blue removal can be achieved to some extent by low cost adsorbent, as some have advantages where by many of them are renewable and available natural resources which are currently under use. Various techniques have been utilized in the removal of dyes. However, practically, a successful methodology for removal of all types of dyes at low cost has not been established.

Antimicrobial activity : Based on the removal efficiency of the methylene blue dye, minimum inhibitory concentration (MIC) was carried out for *Klebsiella pneumoniae* and at different concentrations such as 10 µl, 15 µl and 20 µl for mango seed kernel powder as shown in Fig.5. Antibacterial assay revealed that the mango seed kernel extract showed significant activity against pathogenic bacteria. Antifungal activity with different concentrations was carried out against *Penicillium chrysogenum*. The zone of inhibition is shown in Fig.6. By comparing Fig 5 and Fig.6, it was found that the antimicrobial activity against *Klebsiella pneumonia* and *Penicillium chrysogenum* showed mild enhancement. Hence, it is concluded that using mango seed kernel and Manikara zapota powder was found to be good for removal of methylene blue and basic red dye.

Medicinal plants constitute an effective source of both traditional and modern medicines, but assessment of antimicrobial potential of these sources is essential. The present study found a very promising and readily available source (*M. indica*) for treating infections caused by bacteria

and fungi. This is particularly significant because drug resistance to human pathogens has been increasing not only in the developing countries but throughout the world due to indiscriminate use of antibiotics (Barie *et al.*, 1999).

In a previous research, Vaghasiya *et al.* (2011) found that methanol extract of *M. indica* seeds showed potent antibacterial activity with concentration ranging from 0.6 mg ml⁻¹ to 1.2 mg ml⁻¹. Singh *et al.* (2010) extracted stem bark of *M. indica* and excellent results were obtained with significant antibacterial and antifungal activities (MIC = 0.08 mg ml⁻¹) against *Streptococcus pneumonia*, *Enterobacter aerogenes*, *Klebsiella pneumonia* and *Candida albicans*.

Mango seed kernel powder can be used as an adsorbent for the removal of methylene blue and basic red dyes from its aqueous solutions. The treatment method mentioned above is a chemical free technology and cost effective. When compared to chemical treatment process, the same efficiency of removal could be achieved of this process also. When compared to mango seed kernel powder, mango leaf powder and Manilkara zapota seed powder have achieved - lower dye removal efficiency and that can also be used. For antimicrobial activity, the adsorption techniques are suitable to treat the waste water from dyeing industries. The treatment method mentioned above is a chemical free technology and cost effective. When compared to chemical treatment process, the same efficiency of removal could be achieved by this process also.

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