



Biochar for the removal of pathogens from waste water

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ABSTRACT

Waste water contains various pollutants and pathogens which is harmful to all the living organisms. This waste water without any treatment if released directly to water bodies or soil causes pollution of water and soils. Various agroindustry by-products viz., sawdust, corn cob, wheat husk, bagasse, rice bran, etc. are generated in huge amount which are disposed directly to the landfills or incinerated which causes pollution. The agroindustry by-products can be used to prepare biochar. In this work, wheat husk was used to prepare biochar. This wheat husk based biochar was used for the study of removal of pathogens from waste water. The biochar was also used to check if it can be used for treatment of water by study of total suspended and dissolved solids. The reduction of pathogens in waste water samples A and B using wheat husk based biochar observed was 90 and 80% respectively when compared to untreated waste water samples. Also, no colonies of *E. coli* were observed after treating the waste water samples A and B with wheat husk based biochar. The reduction in total suspended and dissolved solids observed was 97.7 and 76.3% respectively when compared to the untreated water sample. Wheat husk based biochar can be used for the removal of pathogens such as *E. coli* from waste water. The biochar can also be used for the treatment of waste water. The use of biochar for the removal of pathogens, solids and other pollutants from waste water will be very cheap and eco-friendly.

Keywords:

Agroindustry by-Products,
Biochar, Biological,
Economical, Pollutants

1. Introduction

The waste water contains various pollutants such as heavy metals, xenobiotic compounds, solids, pesticide residues, pathogens, etc. [1]. The presence of pathogens such as *E. coli*, *Salmonella*, *S. aureus* sp., coliforms, etc. in the waste water is a serious issue and needs immediate solution [2]. Such waste water without any treatment can be disposed directly to the water bodies or soil which will cause environmental pollution and also harm the living organisms. The removal of pathogens from waste water by chemical approach is costly and causes pollution [3].

Agroindustry by-products such as rice, wheat husk, bagasse, sawdust, corn cob, coconut husk, etc. are generated in huge amount. Some of these agroindustry by-products are disposed directly to the landfills

or incinerated which harms the environment. The agroindustry by-products can be used for the making of 'biochar'. This will also help in the management of agroindustry by-product wastes [4, 5].

The use of biochar for the removal of pathogens from waste water is gaining wide interest [6]. Biochar is charcoal like substance which is produced by pyrolysis by burning agroindustry by-products, agricultural residues, forestry wastes, etc. It is usually black in color and ash-like. Biochar has the adsorption property [7]. The use of biochar for the removal of pathogens from waste water is very eco-friendly, easy and economical. It can thus be used for the treatment of waste water which is essential. Biochar has been found to be excellent for removal of fecal coliform because of the

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adsorption capability [8]. After the removal of pathogens and solids from waste water, such treated water can be used for domestic, agricultural, irrigation, etc. purposes.

The research work here describes the preparation of wheat husk based biochar and use of prepared biochar for the removal of pathogens and treatment of waste water.

2. Materials and methods

2.1 Collection of water samples

The wastewater samples were labeled as A and B. Around 4 l of waste water samples were collected from;

A- Agricultural field Kudale farm, Rahu Taluka, Daund, Pune, Maharashtra, India

B- Well water of Karanje, Satara, Maharashtra, India.

The sample collection sites of waste water shown in Figure 1(i) and (ii) respectively.



Sample A (i)

Sample B (ii)

Fig. 1. Sample collection sites for waste water.

2.2. Agricultural by-Products

The agricultural by-product used for the preparation of biochar was wheat husk. The agricultural by-product was collected from the agricultural field of Karanje, Satara, and Maharashtra, India.

2.3. Processing of the agricultural by-product

The agricultural by-product wheat husk was washed under tap water to remove dust particles (Le *et al.*, 2021) and then kept for drying in the sunlight for two days. After drying, wheat husk was made in powdered form using mixer, packed in clean plastic bag

and labeled properly. The powdered form of wheat husk is represented in Figure 2.

2.4. Preparation of wheat husk based biochar

The processed agricultural by-product wheat husk was placed in clean petridish and kept in the muffle furnace at 550°C for 3 h for the preparation of biochar. The biochar prepared was packed in clean plastic bag and labeled until use. The wheat husk based biochar is shown in Figure 3.



Fig. 2. Powdered form of wheat husk.



Fig. 3. Wheat husk based biochar.

2.5. Study for removal of pathogens from waste water using biochar

2.5.1. Filtration of the waste water samples

The collected waste water samples (50 ml) were filtered using Whatmann filter paper No. 1. The filtrates were collected in clean stoppered bottles and labeled as 'untreated'.

2.5.2. Removal of pathogens from waste water using wheat husk based biochar

Wheat husk based biochar (5 g) was added separately to 50 ml waste water samples A and B respectively and kept for 2 h at room temperature. The samples were used as 'treated with biochar'. After the treatment of waste water with biochar, the total viable count (TVC) was carried. The untreated and treated samples were diluted till 10^{-4} dilution by serial dilution method. 0.1 ml of untreated and treated samples respectively of 10^{-4} dilution was spread on nutrient agar (NA) (g/l) (peptone 10, yeast extract 3, NaCl 5, pH 7.0, agar 30) plates. The plates were kept for incubation in the incubator at 37°C for 24 h and the TVC was determined.

2.5.3. Study of removal of pathogen i.e., *E. coli* from the waste water using the biochar

The MacConkey's agar media was prepared and 0.1 ml of untreated and treated samples respectively of 10^{-4} dilution was spread on MacConkey's agar plates (g/l) (peptone (pancreatic digest of gelatin) 17, proteose peptone 3, lactose 10, bile salts 1.5, NaCl 5, neutral red 0.03, agar 13.5). The plates were kept for incubation in the incubator at 37°C for 24 h and the no. of colonies of *E. coli* was counted on the plates. *E. coli* if present show pink colored colonies due to fermentation of lactose.

2.6. Biochar filter

The wheat husk based biochar filter was made up of four layers separately, first of big stones, second of gravels, third biochar layer which is the main component and fourth of sand. The bacteria present in waste water are removed mainly due to biochar.

2.7. Determination of total suspended and total dissolved solids

For the determination of total suspended (TSS) and total dissolved solids (TDS), the water sample (1000 ml) was collected from Pimpri, Pune, and Maharashtra, India in plastic bottles and labeled properly. The water sample (50 ml) was poured from the top of the biochar filter, which passes the filtration stages. The filtrate was collected in stoppered bottle and was labeled as 'treated'. The water sample without passing through the biochar filter was labeled as 'untreated'. The TSS and TDS were determined using the standard methods of American Public Health Association [9].

3. Results

3.1. Total viable count for untreated waste water samples viz., A and B

The TVC for untreated waste water samples A and B on NA plates is shown in Table 1. The TVC after treating the waste water samples with wheat husk based biochar are shown in Table 1 and 2 respectively.

Table 1. TVC for the untreated water samples

Water sample	No. of colonies / 0.1 ml x dilution factor (10^{-4})	No. of colonies / ml x dilution factor (10^{-4})
A	31	3.1
B	21	2.1

Table 2. TVC after treating the water samples using wheat husk based biochar

Water sample	No. of colonies / 0.1 ml x dilution factor (10^{-4})	No. of colonies / ml x dilution factor (10^{-4})
A	3	0.3
B	4	0.4

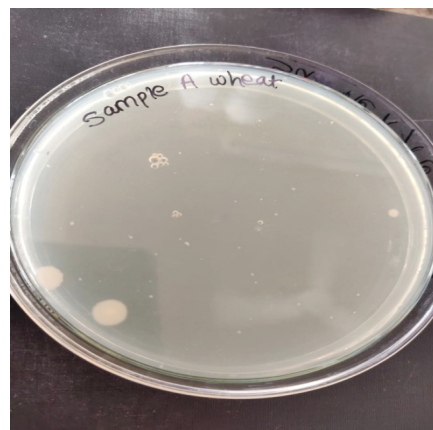
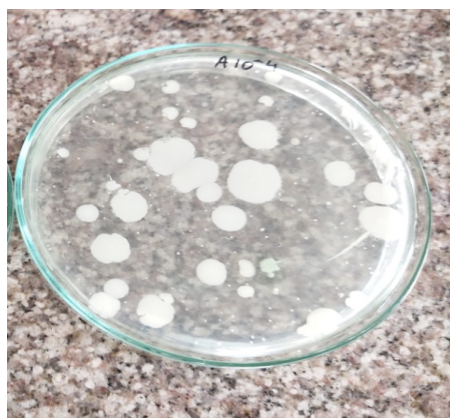


Fig. 4. a- TVC on NA media for the untreated waste water sample A. **b;** TVC on NA media after treating the waste water sample A with wheat husk based biochar.

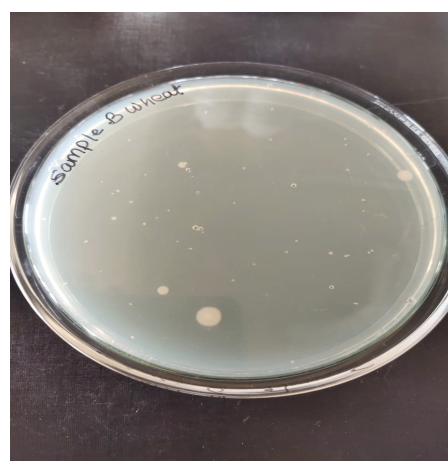
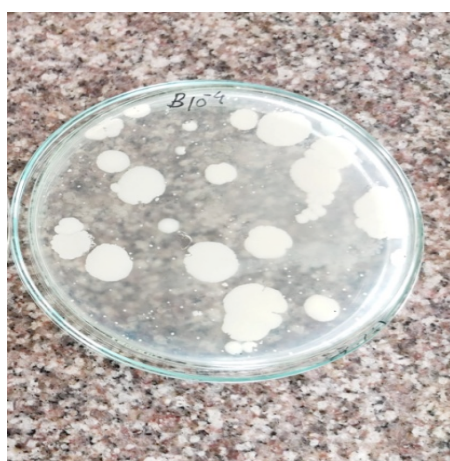


Fig. 5. A; TVC on NA media for the untreated waste water sample B. **b;** TVC on NA media after treating the waste water sample B with wheat husk based biochar.

The no. of colonies is reduced in waste water sample A and B using wheat husk based biochar. In untreated samples A and B, the no. of colonies was 3.1×10^{-4} and 2.1×10^{-4} respectively (Table 1), while in treated water samples A and B using wheat husk based biochar, the no. of colonies was 0.3×10^{-4} and 0.4×10^{-4} respectively (Table 2). The TVC for the untreated and treated water sample A using wheat husk based biochar is represented in Figure 4 a and b respectively. The TVC for the untreated and treated water sample B using wheat husk based biochar is represented in Figure 5a and b respectively.

The reduction of pathogens in waste water samples A and B using wheat husk based biochar was 90 and 80% respectively when compared to the untreated waste water samples.

3.2. Removal of pathogen *E. coli* from waste water using wheat husk based biochar

E. coli colonies on MacConkey agar media were nil after treating the water samples A and B with wheat husk based biochar (Table 3 and 4). *E. coli* colonies on MacConkey's agar media for the untreated water samples A and B are represented in Figure 6a and b. No colonies of *E. coli* were observed on MacConkey agar media after treating the water samples A and B with wheat husk based biochar (Figure 7 a and b).

Table 3. *E. coli* colonies on MacConkey's agar media for untreated water samples

Water sample	No. of colonies / 0.1 ml x dilution factor (10^{-4})	No. of colonies / ml x dilution factor (10^{-4})
A	18	1.8
B	28	2.8

Table 4. *E. coli* colonies on MacConkey's agar media for the treated water samples

Water sample	No. of colonies / 0.1 ml x dilution factor (10^{-4})	No. of colonies / ml x dilution factor (10^{-4})
A	0	0
B	0	0

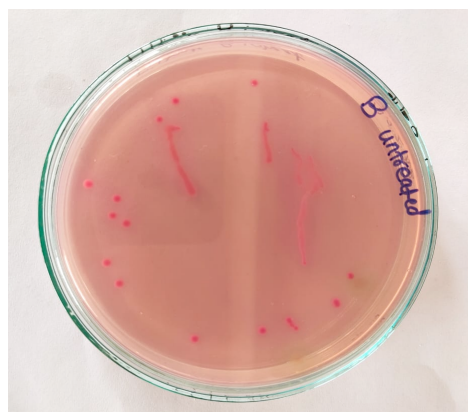


Fig. 6. a; *E. coli* colonies on MacConkey's agar media for the untreated waste water sample A. **b;** *E. coli* colonies on MacConkey's agar media for the untreated waste water sample B.

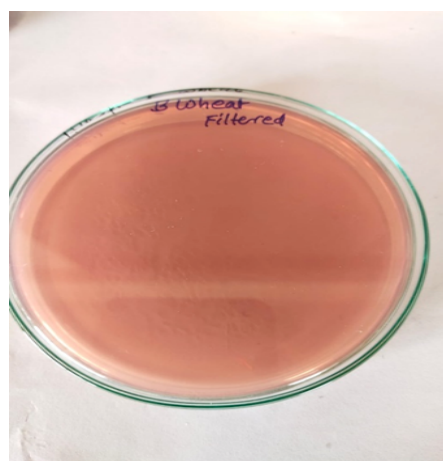
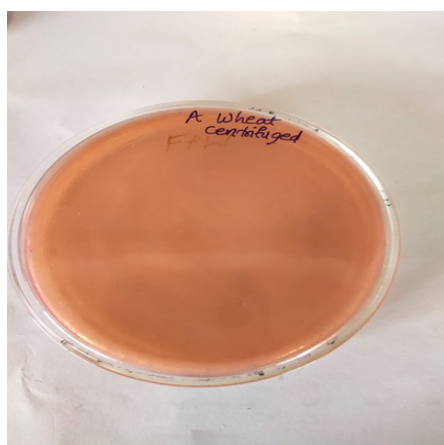


Fig. 7. A; No colonies of *E. coli* seen on MacConkey's agar media after treating the water sample A with wheat husk based biochar. **b;** No colonies *E. coli* seen on MacConkey's agar media after treating the water sample B with wheat husk based biochar.

3.3. TSS and TDS for untreated and treated water sample using wheat husk based biochar

The TSS and TDS for the untreated and treated water sample using wheat husk based biochar are represented in Table 5. The reduction of TSS and TDS was 97.7 and 76.3% respectively when compared to the untreated water sample.

Table 5. TSS and TDS for the untreated and treated water sample using biochar

TSS (mg/l)		TDS (mg/l)	
Untreated water sample	Water sample treated with wheat husk based biochar	Untreated water sample	Water sample treated with wheat husk based biochar
44000	990	4600	1088

4. Discussion

The biochar is effective in removal of pathogens from waste water. There is a report on removal of *E. coli* using biochar modified biofilters [10, 11]. Also study is done where treatment of grey water with biochar filter has minimized the risk of virus infection by 90% [12]. The removal of pathogens from waste water by the biochar depends on the way how it is packed in biofilters. It was reported [6] the use of biochar as a filter for the removal of *Saccharomyces cerevisiae* from waste water. It have studied [13] the removal of bacterial pathogens viz., *Salmonella enterica*, *Staphylococcus aureus*, *E. coli* and MS2 coliphage from stormwater using biochar biofilter. There is a report on use of biochar for the treatment of waste water [14]. There is less work on wheat husk based biochar for removal of pathogens from waste water and treatment of water.

5. Conclusion

The wheat husk based biochar can be used for the clean-up of pathogens such as *E. coli* from waste water. The reduction in TSS and TDS due to treatment of water with wheat husk based biochar indicates the suitability of water for use in agriculture, irrigation and domestic purpose. This technology still needs to be studied on a large-scale for the clean-up of environment. The use of wheat husk based biochar for the removal of pathogens and treatment of waste water will very easy, eco-friendly and economical. The wheat husk agroindustry by-product is also available in huge. Wheat husk is directly disposed to the landfills or incinerated. So, preparation of biochar using wheat husk will also help in the management of this waste.

Conflict of Interests

All authors declare no conflict of interest.

Ethics approval and consent to participate

No human or animals were used in the present research.

Consent for publications

All authors read and approved the final manuscript for publication.

Availability of data and material

All the data are embedded in the manuscript.

Authors' contributions

All authors had equal role in study design, work, statistical analysis and manuscript writing.

Informed Consent

The authors declare not used any patients in this research.

References

1. Haddaoui I, Mateo-Sagasta J. (2021). A review on occurrence of emerging pollutants in waters of the MENA region. *Environmental Science and Pollution Research*, 28: 1-21. <https://doi.org/10.1007/s11356-021-16558-8>
2. Mitiku A A. (2020). A review on water pollution: causes, effects and treatment methods. *Int. J. Pharm. Sci. Rev. Res*, 60(2): 94-101.
3. Seow T W, Lim C K, Nor M H M, Mubarak M F M, Lam C Y, Yahya A, Ibrahim Z. (2016). Review on wastewater treatment technologies. *Int. J. Appl. Environ. Sci*, 11(1): 111-126.
4. Gunjal A B. (2021). Value-Added Products from Agroindustry By-product: Bagasse. In Joshi S J, Deshmukh A, Sarma H (Eds.), *Biotechnology for Sustainable Environment* (pp. 339-351). Singapore: Springer
5. Koul B, Yakoob M, Shah M P. (2022). Agricultural waste management strategies for environmental sustainability. *Environmental Research*, 206: 112285. <https://doi.org/10.1016/j.envres.2021.112285>
6. Perez-Mercado L F, Lalander C, Joel A, Ottoson J, Dalahmeh S, Vinnerås B. (2019). Biochar filters as an on-farm treatment to reduce pathogens when irrigating with wastewater-polluted sources. *Journal of Environmental Management*, 248: 109295. <https://doi.org/10.1016/j.jenvman.2019.109295>
7. Astray G, Fernández-González M, Rodríguez-Rajo F J, López D, Mejuto J C.

- (2016). Airborne castanea pollen forecasting model for ecological and allergological implementation. *Science of The Total Environment*, 548-549: 110-121. <https://doi.org/10.1016/j.scitotenv.2016.01.035>
8. Kaetzel K, Lübken M, Uzun G, Gehring T, Nettmann E, Stenchly K, Wichern M. (2019). On-farm wastewater treatment using biochar from local agroresidues reduces pathogens from irrigation water for safer food production in developing countries. *Science of The Total Environment*, 682: 601-610. <https://doi.org/10.1016/j.scitotenv.2019.05.142>
 9. Chigor V N, Umoh V J, Okuofu C A, Ameh J B, Igbinosa E O, Okoh A I. (2012). Water quality assessment: surface water sources used for drinking and irrigation in Zaria, Nigeria are a public health hazard. *Environmental monitoring and assessment*, 184(5): 3389-3400. <https://doi.org/10.1007/s10661-011-2396-9>
 10. Afrooz A N, Boehm A B. (2016). *Escherichia coli* removal in biochar-modified biofilters: effects of biofilm. *PLoS One*, 11(12): e0167489. <https://doi.org/10.1371/journal.pone.0167489>
 11. Mohanty S K, Cantrell K B, Nelson K L, Boehm A B. (2014). Efficacy of biochar to remove *Escherichia coli* from stormwater under steady and intermittent flow. *Water Research*, 61: 288-296. <https://doi.org/10.1016/j.watres.2014.05.026>
 12. Dalahmeh S S, Lalander C, Pell M, Vinnerås B, Jönsson H. (2016). Quality of greywater treated in biochar filter and risk assessment of gastroenteritis due to household exposure during maintenance and irrigation. *Journal of applied microbiology*, 121(5): 1427-1443. <https://doi.org/10.1111/jam.13273>
 13. Afrooz A N, Pitol A K, Kitt D, Boehm A B. (2018). Role of microbial cell properties on bacterial pathogen and coliphage removal in biochar-modified stormwater biofilters. *Environmental Science: Water Research & Technology*, 4(12): 2160-2169. <https://doi.org/10.1039/C8EW00297E>
 14. Mohan D, Pittman Jr C, Mlsna T. (2022). *Sustainable Biochar for Water and Wastewater Treatment*: Elsevier.



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