

Microbial and Enzymatic Treatment for Decolorization of Distillery Spent-Wash (DSW)



Shivam Kapoor*

Assistant Professor, Environmental Engineering Department, Government Engineering College, India

Submission: August 13, 2018; Published: August 27, 2018

*Corresponding author: Shivam Kapoor, Assistant Professor, Environmental Engineering Department, Government Engineering College, Bhuj, Gujarat, India, Email: shivamkpr88@gmail.com

Mini Review

Being rich in sugarcane yield, all the ethanol, in India, is produced by the way of fermentation of molasses and its subsequent distillation. Sugarcane juice containing sucrose is used by the sugar industry for making sugar. The residue from the sugar-making process, referred as molasses, contains high concentrations of sucrose. Molasses is used as the substrate in fermentors for producing alcohol. The liquor after fermentation contains 8-10 percent ethanol, which is further concentrated and purified in a series of distillation columns. The dark brown opaque liquid remaining after removal of alcohol is disposable and called by various names such as spent-wash, slops, stillage, still bottom, mosto, vinasse and dunder. The distillery spent wash is characterized as one of the caramelized and recalcitrant wastes containing extremely high color, COD, BOD, suspended solids and low in pH. This spent-wash has dark brown color and needs to be decolorized before release into the natural environment. At present, there are 285 distilleries in India that producing 2.7 billion liters of alcohol and generating 40 billion liters of wastewaters annually [1]. Due to high strength of raw spent wash (high biochemical oxygen demand), application of anaerobic treatment technology with biogas recovery has been reported to be highly effective. After anaerobic treatment also effluent contains high concentrations of color and organic pollutants and as such cannot be dispose directly on land and water bodies [2]. The spent wash is highly acidic in nature and has a variety of recalcitrant coloring compounds as melanoidins, phenolics and metal sulfides that are mainly responsible for the dark color of distillery effluent [3].

Nature of Melanoidins

Distillery spent-wash contain melanoidins which are natural condensation products of sugar and amino acids produced by non-enzymatic Millard amino-carbonyl reaction taking place between the amino and carbonyl groups in organic substances. The formation of melanoidins is affected by the reactants and their concentration, type of catalysts and buffers, temperature, time pH, water activity, presence of oxygen and metal ions [4]. Due to complex structure and xenobiotic nature of melanoidins are generally recalcitrant to biodegradation. The empirical formula

of melanoidins has been suggested as $C_{17-18}H_{26-27}O_{10}N$. Disposal of melanoidins rich wastewater reduces sun light penetration leading to decreased photosynthetic reaction and dissolved oxygen concentration in rivers, lakes and lagoons (Figure 1).

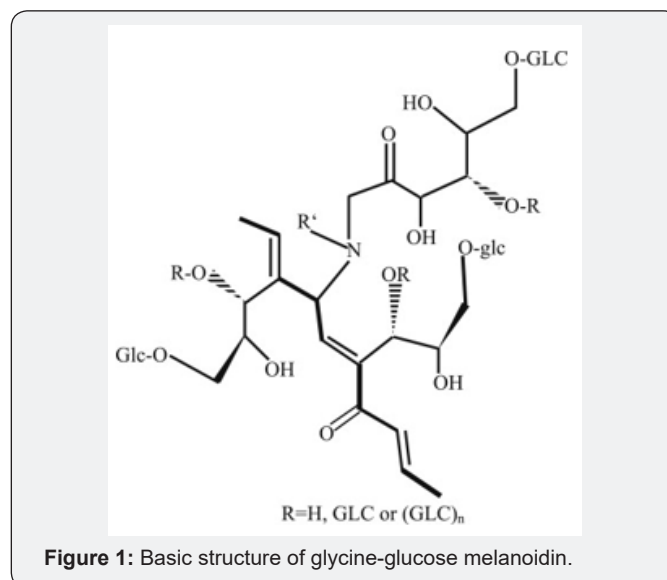


Figure 1: Basic structure of glycine-glucose melanoidin.

Methods for Decolorization

Various physical, chemical, biological and enzymatic processes for decolorization of post anaerobic treatments of spent-wash have been proposed by researchers (Figure 2). In physico-chemical treatment, activated carbon as adsorbent; activated silica, bentonite, polyelectrolytes and starch as coagulant aids; ozone, single hydrogen peroxide and Fenton's reagent as oxidants were used for color removal from distillery spent-wash. All the treatment techniques gave color removal efficiency between 70-90% except single hydrogen peroxide and Fenton's reagent [5]. Although many techniques have been explored for the decolorization but they all are require high reagent dosage and produce large amount of sludge. In biological treatment color can be removed either by concentrating into the sludge or by partial/complete breakdown of color molecules. Anaerobic treated spent wash contains high concentrations of color and organic pollutants.

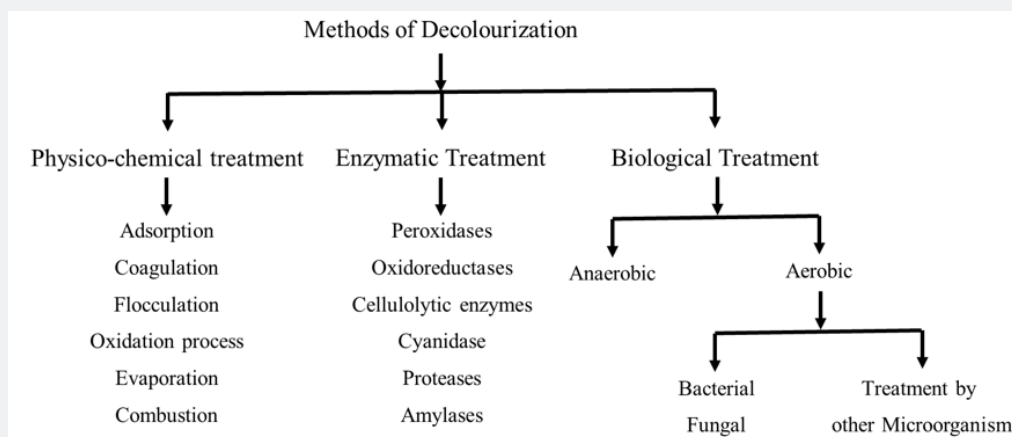


Figure 2: Various methods of decolorization.

Bacterial and Fungal Treatment

Due to the presence of high amount of organic pollutants and formation of toxic products, anaerobically treated wastewater

cannot be directly discharged, and it has to be treated aerobically before discharge. Some of the aerobic based methods are described below Tables 1 & 2.

Table 1: Literature review for bacterial and fungal decolorization.

Culture isolate or Consortium	Initial Conditions	Efficiency	Type of Color Removal Method	Optimized Conditions	Author
Proteus mirabilis, Bacillus sp., Raoultella planticola and Enterobacter sakazakii	MM= 1000 ppm, Temp. = 35°C, 180 rpm, Time= 240 h	75%	Biodegradation (Confirmed by using live and autoclaved cells)	Glucose, peptone, Temp. = 30-35°C, pH = 7.0, Inoculum size = 4:3:2:1, Shaking speed= 100-180 rpm	Yadav and Chandra [11]
Alcaligenes faecalis SAG5	Temp. = 37°C, pH = 7.0, MM= 0.5%, Time= 120 h	72.60%	Not specified	Temp. =37°C, pH= 7.5, Glucose, beef extract	Santal et al. [11]
Pseudomonas aeruginosa PAO1, Stenotrophomonas maltophilia and Proteus mirabilis	Time = 24 h, 0.5% glucose, static, temp = 37°C	67.20%	Not specified	Glucose, Nitrogen source inhibit decolorization, pH= 7, Inoculum= 15%, Temp. = 37°C, Static condition	Mohana et al. [5]
Rhizoctonia sp. D-90	Temp. = 30°C, 125 rpm, 10 ml mycelial suspension, MM= 19.3ppm, time= 192 h, pH= 6, NaOH= 1N	87.5% and 84.5% in molasses medium and synthetic medium	Adsorption on mycelial culture	NaOH concentration= 5N	Sirianuntapiboon et al. [10]
Aspergillus niger 180	Temp. = 30°C, 200 rpm, spores conc. = 10 ¹⁰ spores/L, time= 96 h	69%	83% color removal by biodegradation and 17% by adsorption.	Sucrose (10 g/L), potassium phosphate (1 g/L), phosphate (1 g/L), magnesium salt (100 mg/L), pH= 5.0	Miranda et al. [4]

Table 2: Factors affecting the bacterial decolorization.

Factors	Description
pH	Melanoidins solubility depends on pH; it is less soluble in acidic pH than in alkaline. pH higher or lower than 7.0- 7.5 adversely affected the bacterial growth and degradation capability of the bacterial consortium.
Temperature	Optimum temperature is important for the activity of enzymes and proteins involved in degradation activity. At low temperature the proteins remain inactive while at high temperature they get denatured. Temperature affects the rates of microbial metabolism of melanoidins.

Inoculum Size	Many studies found out that 5%- 7% (w/v) inoculum was the optimum and further increase in inoculum concentration produced more fungal biomass whereas the decolonization of the medium was not improved.
Carbon and Nitrogen Sources	Depending upon the type of bacteria used different carbon sources has been used. Glucose is found satisfactory due to its easy utilization. Similar is the case with nitrogen sources. Mohana et al. [5] reported organic nitrogen at 0.05% (w/v) concentration showed suppressive effect on decolorization and degradation of anaerobically treated distillery effluent.
Shaking Speed	Higher shaking speed above 180 rpm suppressed the decolorization process which might be due to mechanical injury of bacterial cell at high speed

Role of Enzymes in Effluent Decolourization

The enzymatic treatment falls between the physicochemical and biological treatment processes. It has some potential advantages over the conventional treatment. A large number of enzymes (e.g. peroxidases, oxidoreductases, cellulolytic enzymes cyanidase, proteases, amylases, etc.) from a variety of different sources play an important role waste treatment applications. Although the enzymatic system related with decolorization of melanoidins is yet to be completely understood. The white-rot fungi have a complex enzymatic system which is extracellular and non-specific, and under nutrient-limiting conditions is capable of degrading lignolytic compounds, melanoidins, and polyaromatic compounds that cannot be degraded by other microorganisms. Decolorization activity involved two types of intracellular enzymes, sugar-dependent and sugar-independent. Color removal of synthetic melanoidin by *Coriolus hirsutus* involved the participation of peroxidases (MnP and MIP) and the extracellular H₂O₂ produced by glucose-oxidase, without disregard of a partial participation of fungal laccase. The white-rot basidiomycete *T. versicolor* is an active degrader of humic acids as well as of melanoidins. Uniform, small and spongy pellets of the fungus *T. versicolor* were used as inoculum for color removal using different nutrients. Maximum color removal of 82% and 36% removal of N-NH₄ was obtained on using low sucrose concentration and KH₂PO₄ as the only nutrient. The enzyme laccase also helps in decolorizing melanoidins [6-11].

Conclusion

In the few last decades, interest has been developed in the field of bioremediation by using microbes. Several microorganisms such as bacteria and fungi, show a good ability to decolorize the effluent of the melanoidin based distillery industries. Thus, it can be suggested that microbial decolorization holds promise and can be exploited to develop a cost effective, eco-friendly biotechnology package for the treatment of distillery effluent. More technically advanced research efforts are required for searching, exploiting new bacterial species and improvement of practical application to propagate the use of bacteria for bioremediation of industrial

effluents. Broader validation of these new technologies and integration of different methods in the current treatment schemes will most likely in the near future, render these both efficient and economically viable.

References

- Aitken MD, Irvine RL (1989) Stability Testing of ligninase and mnpoxidase from phanerochaete chrysosporium. *Biotechnology and Bioengineering* 34(10): 1251-1260.
- Benito GG, Miranda MP, Santos DR (1997) Decolorization of wastewater from an alcoholic fermentation process with *Trametes Versicolor*. *Bioresource Technology* 61(1): 33-37.
- Cammerer B, Jaluschkov V, Kroh LW (2002) Carbohydrates structures as part of the melanoidins skeleton. *International Congress Series* pp. 1245-1269.
- Miranda PM, Benito GG, Cristobal NS, Nieto CH (1996) Color elimination from Molasses Wastewater by *Aspergillus niger*. *Bioresource Technology* 57(3): 229-235.
- Mohana S, Desai C, Madamwar D (2007) Biodegradation and decolorization of anaerobically treated distillery spent wash by a novel bacterial consortium. *Bioresource Technology* 98(2): 333-339.
- Nandy T, Shastry S, Kaul SN (2002) Wastewater management in cane molasses distillery involving bioresource recovery. *Journal of Environmental Management* 65(1): 25-38.
- Raghukumar C, Mohandass C, Kamat S, Shailaja MS (2004) Simultaneous detoxification and decolorization of molasses spent wash by the immobilized White Rot Fungus *Flavodon flavus* isolated from a marine habitat. *Enzyme and Microbial Technology* 35: 197-202.
- Santal AR, Singh NP, Saharan BS (2011) Biodegradation and detoxification of melanoidin from distillery effluent using an aerobic bacterial strain SAG5 of *Alcaligenes faecalis*. *Journal of Hazardous Materials*, 193: 319-324.
- Santal AR, Singh NP (2013) Biodegradation of melanoidin from distillery effluent: role of microbes and their potential enzymes. *Biodegradation of Hazardous and Special Products* 5: 71-100.
- Sirianuntapiboon S, Sihanonth P, Somachai P, Atthasampunna P, Hayashida S (1995) An adsorption mechanism for melanoidins decolorization by *Rhizoctonia* sp. *Bioscience Biotechnology and Biochemistry* 59(7): 1185-1189.
- Yadav S, Chandra R (2012) Biodegradation of organic compounds of molasses melanoidin (MM) from biomethanated distillery spent wash (BMDS) during the decolorization by a potential bacterial consortium. *Biodegradation* 23(4): 609-620.



This work is licensed under Creative Commons Attribution 4.0 License
DOI: [10.19080/IJESNR.2018.14.555878](https://doi.org/10.19080/IJESNR.2018.14.555878)

**Your next submission with Juniper Publishers
will reach you the below assets**

- Quality Editorial service
- Swift Peer Review
- Reprints availability
- E-prints Service
- Manuscript Podcast for convenient understanding
- Global attainment for your research
- Manuscript accessibility in different formats
(Pdf, E-pub, Full Text, Audio)
- Unceasing customer service

Track the below URL for one-step submission
<https://juniperpublishers.com/online-submission.php>