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Bioremediation Studies of Sugar Mill Effluent by Using Fungal Isolates

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ABSTRACT

The sugar mill effluent was treated with fungal strains such as Penicillium sp., Aspergillus flavus and Aspergillus niger and immobilized fungal consortium as beads were used for the bioremediation. In this study, there was a maximum reduction in BOD and COD values were observed i.e., 56.7% and 62.6% with the effluent inoculated with immobilized culture followed by Aspergillus flavus (40.8%) and Aspergillus niger (50.9%) were reduced the BOD and COD significantly. There was an maximum reduction of total solids (64.4%), total suspended solids (70%) (TSS) and total dissolved solids (64.4%) (TDS) were recorded in sugar mill effluent treated immobilized fungal cultures as compared to the raw effluent which showed 55.5% of total solids, 50% of total suspended solids (TSS) and 56% of total dissolved solids (TDS) on 20 days of bioremediation in sugar mill effluent respectively. A gradual reduction in removal of salinity from 32% to 41.4% as inoculated with different fungal strains and immobilized cultures during 20 days of incubation / bioremediation with sugar mill effluent as compared to untreated effluent (control) showed 27.3% reduction of salinity respectively. The Aspergillus flavus remediated sugar mill effluent showed 34.6% of reduction in salinity was in par with Penicillium species grown in sugar mill effluent (35.9%) on 20th day of incubation respectively.

Keywords : Bioremediation, Sugar mill effluent, Fungal isolates, Physico – chemical parameters, Immobilized beads

I. INTRODUCTION

Sugar mill effluent is generated during the processing of sugarcane in different units such as mill house, boiler house and filters wash. Other than this, improper handling of molasses and this leakage and over flow from storage tanks also contribute to high pollution load [1] . Generally sugar mill effluent is lightly blackish ash in colour with disagreeable

odour, high value of BOD, COD and total suspended solids.

Waste water from sugar industries, if not treated properly, contains significant amount of TDS and TSS. This water may not be useful for crop land irrigation. There are reports which indicate that infiltration rate decreases with increased loading of BOD and TDS and TSS. The high value of TSS can cause decrease in soil porosity due to salt deposition. High TDS value in waste water may also have adverse effect on crops [2]. A TDS of 500-1000 ppm may have detrimental effect on sensitive crops.

II. RELATED WORK

Growth rate of various fungal species inoculated with the sugar mill effluent also observed throughout the study and their increase in growth was independent upon the nature of fungus. The COD value is usually higher than the BOD because some organic materials in the water that are resistant to microbial oxidation and hence not involved in BOD could be easily chemically oxidized [3].

Therefore in this study an attempt has been made to bring out the capabilities of fungi for bioremediation of sugar mill effluent and the efficiency of bioremediation was finally validated with the selective fungal isolates as native organism based on the values obtained for the parameters such as pH, TDS, BOD, COD and salinity.

III.MATERIALS AND METHODS

Initial physico - chemical parameters like colour, odour, pH, total suspended solids (TSS), total dissolved solids (TDS), biological oxygen demand (BOD), chemical oxygen demand (COD) and salinity were measured [6].

Effluent collected from the sugar mill was autoclaved at 121^0C for 15 minutes to make them sterile before inoculating the selected fungal strains. To this, 10^6 cells / ml of the uniform suspension of each fungal strain was inoculated into 100ml of sterile sample taken in Erlenmayer flasks. Then it was covered with cotton and various parameters were measured regularly at every five days interval.

Preparation of Immobilized Fungal Consortium:

The fungal consortium (fungal isolates) were immobilized as beads according to the procedure [7] in which two percent sodium alginate solution was prepared in sterile distilled water by heating it to 60^0C and mixing it thoroughly on a magnetic was cooled at room temperature and 10% (10ml culture in 100ml Sodium alginate solution) of the cell culture was added. The contents were mixed well by vigorous shaking to get a homogenized mixture.

In a separate beaker, 100ml of 0.1M calcium chloride solution was taken. The sodium alginate containing cell culture suspension was extracted drop wise through a syringe and allowed to fall in the beaker containing calcium chloride solution. The beads of sodium alginate gel formed are left in the beaker overnight for hardening. Then beads were washed and stored in distilled water at 28±2°C.

Bioremediation of sugar mill effluent using immobilized fungal consortium:

The collected sugar mill effluent was inoculated with immobilized beads 5% inoculum containing of fungal consortium and incubated at 30 ± 2°C. The sample was filtered under aseptic conditions and physico chemical parameters (pH, TSS, TDS, BOD, COD and salinity) were estimated. Measurements were done with control also.

Dissolved oxygen (DO) was measured using the modified winklers method and biochemical oxygen demand (BOD) with the five day incubation method. Chemical oxygen demand was carried out using the potassium permanganate method. Salinity was determined using Mohr's method where chromte ions as an indicator in the titration of chloride ions

with a silver nitrate as standard solution. pH was measured with ELICO analytical pH meter model Li-15. TSS and TDS were determined by the standard procedure [8] .

IV. RESULTS AND DISCUSSION

Sugar mill effluent is generated during the processing of sugarcane in different units such as mill house, boiler house and filters wash. Other than this, improper handling of molasses and this Leakage and over flow from storage tanks also contribute to high pollution load [1]. Generally sugar mill effluent is slightly blackish ash in colour with disagreeable odour, high value of BOD, COD and total suspended solids [9]. The physico- chemical characteristics of the collected sugar mill effluent was analyzed and the results were showed in Table - 1.

Some potential fungal strains such as *Penicillium sp., Aspergillus flavus* and *Aspergillus niger* were isolated from sugar mill effluent and identified based on colony characters. These fungal strains were immobilized as fungal consortium as beads and used for the bioremediation of the collected sugar mill effluent. Similar findings are supported where the life in effluent is highly diverse and consists of interacting population of micro organisms and effluent fauna and their activities affect physical, chemical and biological characteristics of effluent [10,11].

There was a drastic reduction in pH of effluent from initial day to 20th day as bioremediated with various fungal isolates (*viz*., *Penicillium sp., Aspergillus flavus* and *Aspergillus niger*).

The immobilized fungal strains grown in effluent showed a decline in pH from 10.7 to pH 7.1 upto 20th days of incubation respectively (Table 2). This pH reduction was gradually observed in *Aspergillus niger, Aspergillus flavus* and *Penicillium* inoculated with effluent as compared to control (pH 7.8).

With the inoculation of different fungal strains and immobilized culture to the sugar mill effluent, there was a decrease in pH, COD, BOD, TDS and TSS as compared to raw/untreated effluent till / upto 20 days of bioremediation respectively. Similar findings which were recorded in the present study was also reported by various researchers as variations in physico - chemical properties may be due to the processed involved raw materials used and chemicals used in the sugar mill [12, 13,14 ,15].

After treatment with fungal isolates, the effluent turned colourless and odourless, which might be due to the action of fungal isolates [16]. The pH of the effluent was basic prior to biotreatment. Later on the pH was changed to neutral due to bioremediation of the effluent. But, the pH of the effluent was acidic prior to biotreatment, this was contradictory/negative to our result [17].

There was a significant reduction in BOD and COD values were observed *i.e*., 56.7% and 62.6% with the effluent inoculated with immobilized cultures of bioremediation. The percentage of reduction in BOD and COD were 40.8% by *Aspergillus flavus* and 50.9% by *Aspergillus niger*. The fungal species *Penicillium* brought a reduction in BOD and COD to extend of 39.8% and 44.5% as compared to control (untreated) sample showed a reduction of 41.2% in BOD and 32.% in COD on 20th day of incubation with sugar mill effluent respectively. The percentages of reduction for all these parameters were collectively showed in Table - 3 and 4. The process of bioremediation was generally understood with higher percentage of reduction accompained with BOD and COD. Our results are positively correlated in accordance with Buvaneswari *et al*. (2013). These values were noted in control as well as for samples of sugar mill effluent inoculated with different fungal strains and immobilized fungal cultures [9] .

Growth rate of various fungal species inoculated with the sugar mill effluent also observed throughout the study and their increase in growth was independent upon the nature of fungus. The COD value is usually higher than the BOD because some organic materials in the water that are resistant to microbial oxidation and

hence not involved in BOD could be easily chemically oxidized [3] .

There was a gradual reduction in removal of salinity from 32% to 41.4% as inoculated with different fungal strains and immobilized cultures during 20 days of incubation / bioremediation with sugar mill effluent as compared to untreated effluent (control) showed 27.3% reduction of salinity respectively. The *Aspergillus flavus* remediated sugar mill effluent showed 34.6% of reduction in salinity was in par with *Penicillium* species grown in sugar mill effluent (35.9%) as shown in Table. 5.

The total dissolved solids (TDS) and total solids (TS) of raw sugar mill effluent (untreated) were 2500 and 2700 mg/l. When the sugar mill effluent was treated with different fungal cultures as monoculture and immobilized fungal consortium, showed a drastic reduction in TDS and TS as bioremediated for 20 days respectively. There was an maximum reduction of total solids (64.4%), total suspended solids (70%) (TSS) and total dissolved solids (64.4%) (TDS) were recorded in sugar mill effluent treated immobilized fungal cultures as compared to the raw effluent which showed 55.5% of total solids, 50% of total suspended solids (TSS) and 56% of total dissolved solids (TDS) on 20 days of bioremediation in sugar mill effluent respectively (Table 6 and 7).

Several scientists were reported that TDS was also found to be high before treatment of sugar mill effluent which may be due to the presence of high salt content and in bioremediated effluent it showed reduction indicating that the microbial isolates have the efficiency to reduce the TDS in the sugar mill effluent and also fungi *Phanerochaete chrysosporium* [18,19,20,21] .

Penicillium where as among the monoculture of fungal isolate treated effluent showed less reduction of total solids (58%), total suspended solids (50%) and total dissolved solids (52%) by *Aspergillus flavus* in 20 days of bioremediation respectively (Table 8).

These findings were also in accordance to various researchers observed that rate of dissolved colliding particles is referred to collision and the pH affected dissolved rate of collision in the effluent [9,22,23,24] . Dissolved and non-dissolved substances called as total solids and it composed of carbonates, chlorides, sulphides, bicarbonates, nitrates [24] .

In recent years, several basidiomycetes and ascomycetes type of fungi have been used in the decolouration of natural and synthetic melanoidin in connection with color reduction of waste waters from distilleries [11] .

The fungus have capability to purify the effluent by consumption of organic substances, thus reducing its COD and BOD and at the same time to obtain some valuable product, such as fungal biomass for protein - rich animal feed or some specific fungal metabolite.

Similar findings are in positively correlated that a single microbe do not have all the enzymes to degrade different kinds of organic matter. Therefore a mixture of microbes of various kind is required to completely degrade the organic matter which justifies the present investigation. Hence, it may be suggested that instead of using monoculture of fungus, co culture / mixed cultures with immobilization will be beneficial for biodegradation and purification of effluents [19].

V. CONCLUSION

Mechanism of microorganisms in control of environmental pollution is still being explored. However, it is argued that organisms during bioremediation either eat up / gobble the contaminants especially organic compounds / assimilate heavy metals themselves, thus effectively degrading specific contaminants / harmful compounds and converting them to non - toxic useable by products.

This study concluded that physico - chemical parameters such as pH, TSS, TDS, BOD, COD and

salinity were relatively high in the sugar mill effluent and severely affected the environment and water bodies. The sugar industry effluent which is untreated highly toxic to plants and it is not permissible for irrigation. The immobilized fungal consortium and individual isolated fungal orangisms (*Aspergillus niger, Aspergillus flavus* and *Penicillium*) were used for the bioremediation of sugar mill effluent and showed a drastic reducton in the levels of COD, TSS, TDS at every 5 days interval respectively.

VI. FUTURE SCOPE

Biotreatment offers easy, effective, economical and ecofriendly techniques and utilization of immobilized fungal consortium, also mono cultures of fungal isolates can be applied for fine tuning of sugar mill effluent treatment. The treated effluents of industry are not highly polluted and therefore can be used for irrigation purpose and also has the potentiality as organic fertilizer.

VII. CONFLICT OF INTEREST

The authors hereby declare that they had no conflict of interest

VIII. REFERENCES

[1]. Rao, M.N. and A. K. Dutta 1987. Waste water treatment Second edition, Oxford and Publishing company Pvt. Ltd.

[2]. Mohana , S., C. Desai and D. Madamwar, 2007. Biodegradation decolorisation of an aerobically treated distillery spent wash by a novel bacterial consortium . Biores. Technol. 98: 333 – 339.

[3]. Aniyikaiye, E. T., T. Oluseyi and Joshua N. Edokpayi, 2019. Physico chemical Analysis of Wastewater Discharge from Selected Paint Industries in Lagos, Nigeria. Int. J. Environ. Res. Public Health, 16, 1235; doi:10.3390/ijerph16071235w.

[4]. Booth, E. 1971. Methods in microbiology, Academic Press. New York.

[5]. Onions, A.H.S., D.Allsopp, and H.O.W. Egginns 1981. Smiths Introduction in Industrial Mycology (7th ed). Edward Arnold Publishers Ltd.

[6]. APHA . 2005. Standard methods for the examination of water and wastewater, 21st edn. American Public Health Association, Washington.

[7]. Leung, W.C., M.F. Wong and C.K. Leung 2000. Removal and recovery of heavy metals by bacteria isolated from activated sludge treating industrial effluents and municipal waste water. Water Science Technology, 12: 233 - 240.

[8]. Richard, L. A. 1954. Diagnoses and improvement of saline and alkali soils. Agriculture Hand Book , 60: USDA, USA.

[9]. Buvaneswari.S., S. Damodarkumar and S. Murugesan 2013. Bioremediation studies on Sugar mill effluent by selected fungal species. Int J Curr Micorbial Appl Sci., 2(1): 50 – 58.

[10]. Pant , D. and A. Adholeya 2007b. Biological approaches for treatment of distillery waste water: a review. Bioresource Technoloy , 98, pp 2321 – 2334.

[11]. Jai Shanker P., N. Danesh, E.T.Puttaiah and K.Girish 2011. Microbial diversity in solid waste molasses of Sugar Industry, Aranthangi, Tamilnadu . International journal of environmental sciences, 2 (2) ; 723 - 30.

[12]. Lakshmi. S. and A. Sundaramoorthy 2002. Effect of Sugar mill effluent on germination, seedling growth and biochemical changes in Ragi. Indian Journal Environment and Ecoplant, 3(3): 501 – 506.

[13]. Rathore .N .P., S.A. Iqbal and K.S. Pawan 2000. Role or Sugar Industry effluent in agriculture. Indian J Appl Pure Biol ., 19: 91 – 94.

[14].Thamizhiniyan. P., P.V. Sivakumar, M. Lenin and M. Sivaraman. 2009. Sugar mill effluent toxicity in crop plants. Journal of Phytology, 1(2): 68 - 74.

[15].Borole .D.D. and P.R. Patil 2004. Studies on physicochemical parameters and concentration of heavy metals in sugar industry. Pollut. Res., 23: 83 – 89.

[16].Glick, B.R. 2003. Phytoremediation: synergistic use of plants and bacteria to clean up the environment. Biotechnology Advances, 21: 383 – 393.

[17].Saranraj . P. and D. Stella 2012. Effect of bacterial isolates on reduction of physic – chemical characteristics in sugar mill effluent. Int J Pharm Biol Arch., 3 (5): 1121 – 1128.

[18].Jesudass, L and R. Akila. 1996. Studies on the physico-chemical characteristics of the sugar factory effluent. Internationa Journal of Ecobiotechnology, 16 (11): 65 - 67.

[19].Goel, P.K. 2000. Water pollution causes, effects and control. New Age International (P) Lts., Publisher. New Delhi, p 269.

[20].Devarajan L., G. Rajanan and P.P Ramaswami. 2001. Bio earth composting: a technology for recycling of sugar and distillery wastes. Final Report. Tamil Nadu Agricultural University. P 44.

[21].Prabakar, K., S. Dawood Sharief and Nausheen Dawood. 2010. Bioremediation of sugar mill effluent using white rot fungus, Phanerochaete chrysosporium. Italian Journal of Applied Biology,1(1) : 31 - 35.

[22].Vinod ,K., and A.K. Chopra 2014a. Fertigational effects of Sugar mill effluent on agromonical characteristics of high yield cultivar of sugarcane (Saccharum officinarum L.) in two seasons. Acta Adv Agric Sci ., 2 (9) : 17 – 39.

[23].Vinod K and A.K. Chopra 2014b. Pearl millet (Pennisetum glacicum L.) response after ferti – irrigation with sugar mill effluent in two seasons. Int .J. Recycl .Org Waste. Agric 3: 67. https://doi.org/co.1007/340093-014-0067.

[24].Vaithiyanathan . T. and P. Sundaramoorthy 2017.Analysis of sugar mill effluent and its influence on germination and growth of African marigold (Tagetes erecta L.). Appl Water Sci ., 7:4715 – 4723.

Table.1: Physico- chemical Properties of sugar mill effluent

S.No.	Parameters	Standard by TNPCB(2009)	Values
1	Colour	Colourless	Light Brown
2	Odour	Odourless	Odourless
3	pH	5.5 - 9	9.72
4	Total Suspended solids(mg/l)	200	200
5	Total Dissolved Solids(mg/l)	200	250
6	Biological Oxygen Demand (mg/l)	30	980
7	Chemical Oxygen Demand (mg/l)	250	1989
8	Salinity (mg/l)	600	780

- TNPCB- Tamil Nadu Pollution Control Board

Table 2. pH Reduction in bioremediated sugar mill effluent using fungal isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	9.5	9.01	8.23	7.8
2	Sugar mill effluent + *Aspergillus niger*	8.9	8.1	7.7	7.3
3	Sugar mill effluent + *Aspergillus flavus*	8.9	8.3	7.6	7.23
4	Sugar mill effluent + *Penicillium*	9.23	8.7	8.07	7.62
5	Sugar mill effluent + Immobilized culture	9.01	8.5	7.62	7.1

Table 3. Removal efficiency of COD (%) in Bioremediated Sugar mill effluent using fungal isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	17.5	30.4	35.8	41.2
2	Sugar mill effluent + *Aspergillus niger*	3.2	17.5	33.2	50.9
3	Sugar mill effluent + *Aspergillus flavus*	7.64	27.6	33.2	47.5
4	Sugar mill effluent + *Penicillium*	3.2	13.1	42.7	44.5
5	Sugar mill effluent + Immobilized fungal cultures	20.7	37.2	40.9	62.6

Table 4. Removal efficiency of BOD (%) in bioremediated Sugar mill effluent using fungal Isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	10.2	22	28.6	32.6
2	Sugar mill effluent + *Aspergillus niger*	18.4	25.1	29.5	34.7
3	Sugar mill effluent + *Aspergillus flavus*	20.4	27.5	35.7	40.8
4	Sugar mill effluent + *Penicillium*	18.4	24.5	33.6	39.8
5	Sugar mill effluent + Immobilized fungal cultures	22.4	36.7	45.5	56.7

Table. 5. Salinity reduction in bioremediated Sugar mill effluent using fungal isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	5.1	13	23	27.3
2	Sugar mill effluent + *Aspergillus niger*	10.5	15.4	25.4	32
3	Sugar mill effluent + *Aspergillus flavus*	7.7	17.3	26.6	34.6
4	Sugar mill effluent + *Penicillium*	8.3	20.5	30.1	35.9
5	Sugar mill effluent + Immobilized fungal cultures	8.9	19.2	34.6	41.4

Table : 6. Reduction in Total Dissolved Solids (TDS) (%) of bioremediated sugar mill effluent using fungal Isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	12	24	40	56
2	Sugar mill effluent + *Aspergillus niger*	16	28	44	60
3	Sugar mill effluent + *Aspergillus flavus*	8	32	44	52
4	Sugar mill effluent + *Penicillium*	4	20	36	52
5	Sugar mill effluent + Immobilized fungal Consortium	20	36	48	64

Table : 7. Reduction in Suspended Solids (SS%) in Bioremediated Sugar mill effluent using fungal isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	12	20	40	50
2	Sugar mill effluent + *Aspergillus niger*	15	32.5	45	60
3	Sugar mill effluent + *Aspergillus flavus*	20	30	37.5	57.5
4	Sugar mill effluent + *Penicillium*	10	27.5	35	50
5	Sugar mill effluent + Immobilized fungal Culture	25	40	55	70

Table 8. Removal of Total Solids (TS%) in Bioremediated Sugar mill Effluent using fungal isolates

S. No.	Samples	Days of Bioremediation			
		5	10	15	20
1	Sugar mill effluent	11.8	10	15	20
2	Sugar mill effluent + *Aspergillus niger*	16	28.3	44	60
3	Sugar mill effluent + *Aspergillus flavus*	8.8	31.8	43.5	59.4
4	Sugar mill effluent + *Penicillium*	4.4	20.5	35.0	51.8
5	Sugar mill effluent + Immobilized fungal Culture	20.37	36.3	48.5	64.4

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