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### **A Study on the Potential of *Aspergillus Flavus* in Degradation of Coffee Processing Effluent**

***K R Navitha\* and Kousar Hina***

<sup>1</sup>Department of PG Studies & Research in Environmental Science, Kuvempu University, Shankaraghatta, 577451. Shivamogga, Karnataka. India

#### **ABSTRACT**

An investigation was carried out to assess the potential of *Aspergillus flavus* in reducing certain physico-chemical parameters of the coffee processing effluent such as total suspended solids (TSS), total dissolved solids (TDS), biological oxygen demand (BOD) and chemical oxygen demand (COD). *Aspergillus flavus*, was isolated from the coffee processing effluent was introduced into the waste water. The initial concentration of the above-mentioned parameters in the effluent was found to be very high, which drastically reduced upon introduction of *A.flavus*. The reduction efficiency of *A.flavus* was observed for different concentrations i.e. 25%, 50%, 75% and raw effluent. It was observed that maximum reduction was found in 25% dilution. Total suspended solids reduced by 94%, total dissolved solids by 86%, biological oxygen demand by 89%, and chemical oxygen demand by 79% there by proving that *A.flavus* is highly potent in reducing the concentration of the effluent.

**KEY WORDS:** Coffee processing effluent, Physico- chemical parameters, *A.flavus*.

#### **\*Corresponding author\***

***K R Navitha***

Research Scholar,

Department of Environmental Science, Kuvempu University,  
Shankaraghatta, Shimoga, Karnataka, India.

Email: [navithaevs29@gmail.com](mailto:navithaevs29@gmail.com), [eshinakousar@gmail.com](mailto:eshinakousar@gmail.com)

## INTRODUCTION

Coffee is one of the main commercial crop cultivated in southern parts of India. Coffee pulping plants are mainly situated along the banks of rivers and streams. Coffee processing requires large quantity of water for wet processing of cherries. Coffee pulping plants release effluent in large amounts with its attendant polluting property. The wastewater is acidic, consisting of high concentration of organic matter, nutrients and suspended matter, which leads to a very high pollution load<sup>7, 10</sup>. The rise in the number of wet processing plants has therefore resulted in the generation of large amounts of processing by-products<sup>11</sup>. Coffee processing plants have adverse environmental effects including surface and ground water pollution. The introduction of contaminants through effluent and sludge to different water bodies can often overcome the self-purifying capacity of recipient ecosystems and thus result in the accumulation of pollutants to noxious levels<sup>2</sup>. Thus, coffee processing plants require effective treatment to avoid contamination to receiving water bodies. The biological treatment of coffee effluent depends primarily on consortium of microbial activities, which uptake organic substances present in the effluent as supplements and degrade.

## MATERIALS AND METHODS

**Effluent Source:** The sample for the analysis was collected from a coffee processing unit in Chikmagalur District of Karnataka, India and stored at 4°C in the laboratory.

**Isolation and Identification of microorganisms:** The sample was serially diluted using sterile pipettes from 10<sup>-1</sup> to 10<sup>-8</sup> dilution. The isolated fungal culture was identified as *Aspergillus flavus* using Lactophenol cotton blue staining method based on their morphology and reproductive structural characteristics<sup>6</sup>. Isolated pure cultures were maintained on PDA media and stored at 4°C.

**Experimental Design:** The effluent was diluted to different concentrations viz. 25%, 50%, 75% and raw effluent is designated as 100% concentration. The effluent was filtered through Whatman No 1 filter paper and transferred to 250ml conical flask which is supplemented with 0.5% concentration of glucose to improve the degradation efficiency of the microorganisms<sup>2</sup>. The nine day's old isolated *Aspergillus flavus* was inoculated into the conical flask. The conical flask containing effluent without introducing *Aspergillus flavus* was kept as control. The anaerobic treatment under aseptic condition was maintained in laboratory scale and treatment was conducted for a period of seven days in triplicates. Physicochemical parameters like TSS, TDS, BOD, and COD were analysed before and after treatment. The percentage reduction of the treatment process for different concentration was calculated using the following equation,

$$\text{Reduction \%} = \frac{C_i - C_f}{C_i} \times 100$$

Where,  $C_i$  is the concentration of the coffee effluent before treatment and  $C_f$  is concentration of the coffee effluent after treatment

**Data Analysis:** Statistical analysis was carried out using one-way ANOVA followed by Tukey's t-test. The difference in mean values at  $p < 0.05$  or  $p < 0.01$  were considered as statistically significant. Statistical analysis was performed using ez ANOVA 0.98 version.

## RESULTS AND DISCUSSION

The concentration of physico-chemical parameters viz TSS, TDS, BOD and COD after treatment is enumerated in Table 1, results are expressed as mean $\pm$ SD which is statistically significant. It is observed that concentration of total suspended solids after treatment was reduced by 84% in raw effluent, 88% in 75% concentration, 92% in 50% concentration, 94% in 25% concentration and 12% in control. Total dissolved solids (mg/L) reduced gradually by 78.46% (raw effluent), 81.81% (75% concentration), 84.71% (50% concentration) and 85.63% (25% concentration) whereas in control 15.04% reduction of total dissolved solids was observed(Figure.1).*Aspergillus flavus* reduced BOD to 82.44% (raw effluent), 83.85% (75% concentration), 86.95% (50% concentration) and 89.40% (25% concentration) where as in control, 16.71% reduction was obtained. From Figure 1, it is observed that percentage of BOD removal increased with increase in dilution of the effluent. This reveals that dilution increases the efficiency of biological degradation. COD (mg/L) in the effluent reduced by 74.14% (raw effluent), 75.78% (75% concentration), 77.37% (50% concentration), 79.49% (25% concentration) and in control COD was reduced to 10.34%. It is evident from Figure 1 that after treatment maximum reduction of all the listed parameters was seen in in 25% concentration. Concentration of parameters has reduced significantly with increasing dilution, because dilution enhances biological degradation and fermentation.

**Table 1: Concentration of the effluent parameters after treatment with *Aspergillus flavus*.**

Parameters	Control	Raw	75% Concentration	50% Concentration	25% Concentration
TSS(mg/L)	2563.66 $\pm$ 2.51	467.23 $\pm$ 0.251**	340.23 $\pm$ 0.32**	190.5 $\pm$ 0.45**	113.33 $\pm$ 0.3**
TDS(mg/L)	2710.4 $\pm$ 0.45	687.23 $\pm$ 0.25**	540.23 $\pm$ 0.32**	390.5 $\pm$ 0.45**	333.33 $\pm$ 0.3**
BOD(mg/L)	3748.26 $\pm$ 0.2	790.17 $\pm$ 0.2**	630.08 $\pm$ 0.1**	339.31 $\pm$ 0.35**	150.44 $\pm$ 0.5**
COD(mg/L)	5824.23 $\pm$ 0.25	1680.08 $\pm$ 0.1**	1394.31 $\pm$ 0.35**	970.22 $\pm$ 0.25**	590.043 $\pm$ 0.05**

Key: (mg/L) = milligram per litre. Values are expressed as mean  $\pm$  SD (n=3), \* $p < 0.05$ ; \*\* $p < 0.01$ , denotes significance with respect to initial values using one-way ANOVA followed by Tukey's test.

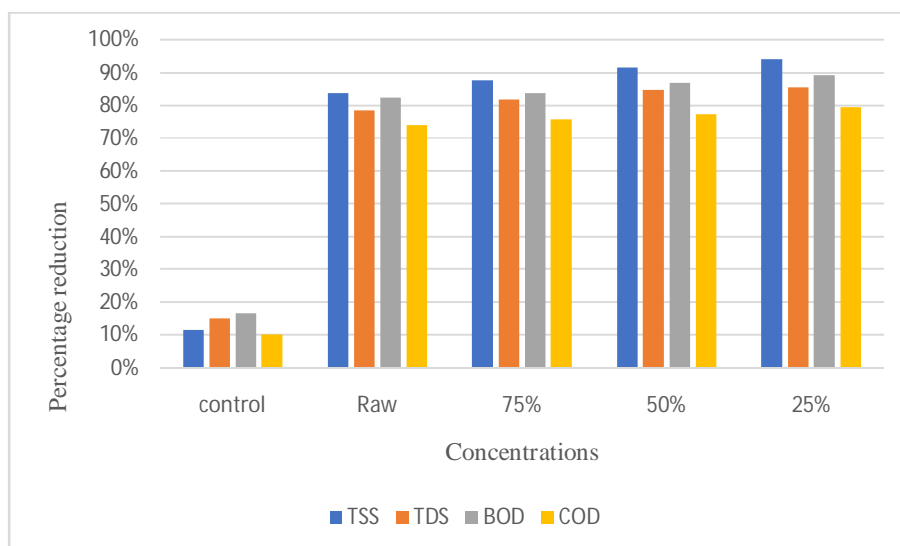


Figure 1: Percentage reduction of effluent parameters after treatment with *Aspergillus flavus*.

## CONCLUSION

The current study elucidated that the anaerobic degradation of the organic contaminants of coffee effluent could be carried out by inoculating with fungus *Aspergillus flavus* under optimal conditions. Microbial degradation can effectively reduce all the observed parameters in a short degradation period. Maximum reduction of parameters was observed in 25% concentration of the effluent.

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