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Biodegradation of Dianix Yellow Brown Azo Dye by *Paramecium jenningsi* Isolated from Industrial Wastewater

Tuba Arooj, Itrat Zahra, Farah Rauf Shakoori
[1]Institute of Zoology, University of the Punjab, Quaid-i-Azam Campus, Lahore, Pakistan

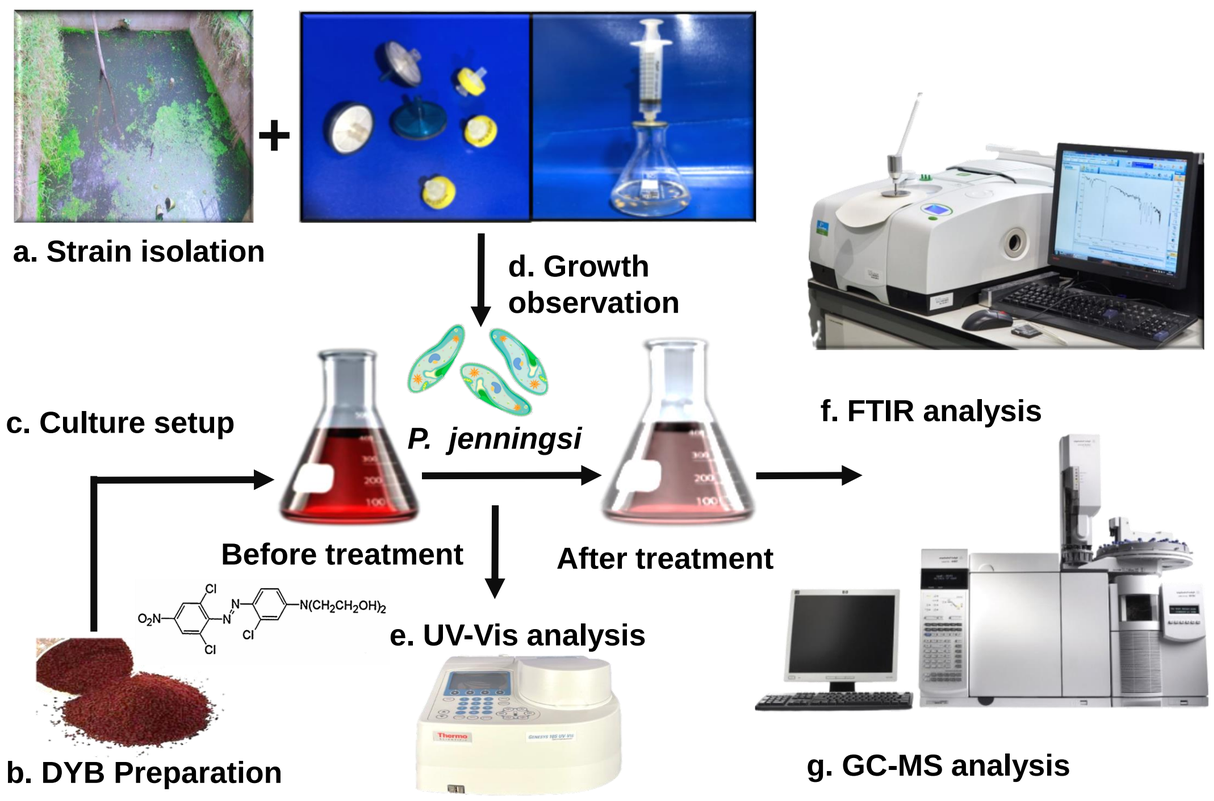
INTRODUCTION & AIM

Introduction: Synthetic dyes are produced in over 100,000 forms, with ~7 × 10^5 tons made annually. The textile sector uses two-thirds of these dyes and discharges ~280,000 tons into wastewater, contributing ~20% of industrial water pollution [1,2]. Azo dyes, the dominant class (80% of textile dyes), are cheap and stable but reduce light penetration, hinder photosynthesis, raise oxygen demand, and release toxic, mutagenic intermediates in aquatic environment. Conventional treatments are costly and often incomplete, whereas bioremediation offers an eco-friendly alternative using microorganisms to convert dyes into less harmful products [2,3]. Recent studies highlight ciliated protozoa (*Paramecium* sp.) as promising agents for azo dye removal due to their adaptability and xenobiotic metabolism [4].

Aim: This study investigates the potential of an industrial wastewater strain of *Paramecium jenningsi* to degrade Dianix Yellow Brown (DYB), a widely used but poorly studied polyester azo dye, highlighting a novel approach for sustainable wastewater treatment.

METHOD

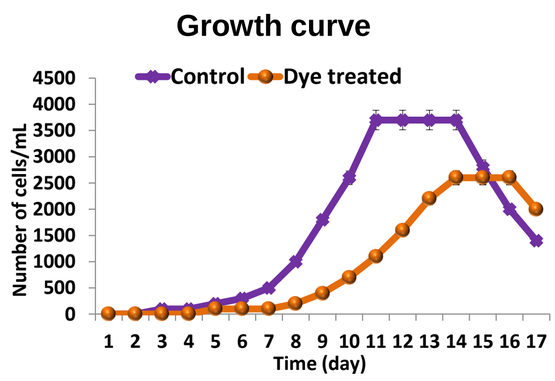
1. Culture and Dye Preparation: DYB (≥95% purity) was purchased from Sigma-Aldrich, prepared as a 0.1% stock solution, and then diluted to a 20ppm working solution. *P. jenningsi* was isolated from industrial wastewater and identified by 18S rRNA sequencing (NCBI Accession No. MZ540265). Cultures were maintained in Bold's Basal Medium (BBM) with boiled wheat grains, and daily growth monitoring was performed under a compound microscope.



2. Experimental Procedures: Optimum conditions for *P. jenningsi* under DYB stress (20 ppm) were determined (pH 6–8.5; 20–35 °C). At these conditions, growth, morphology, motility, and vacuole activity were monitored. Decolorization was measured by UV–Vis (260 nm); FTIR and GC–MS analyzed structural changes and metabolites. Experiments were conducted in triplicate; data analyzed as mean ± SD (ANOVA; DMRT, $p < 0.05$).

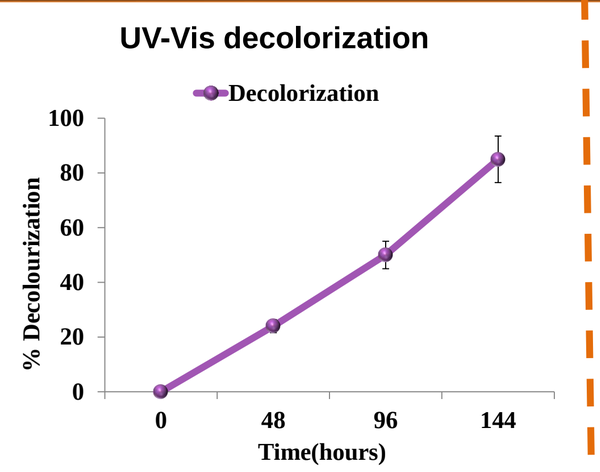
RESULTS & DISCUSSION

Under optimum conditions, pH 7 and temperature 25°C, *P. jenningsi* showed ~30% reduced growth with marked declines in mitosis, motility, and vacuole activity under DYB stress.



Days	Morphological and behavioral changes		
	Shape	Speed	Vacuole
5th	Oval elongated	Slightly slow	Slightly darker
11th	Swallowed elongated	Very Slow	Darkest
17th	Oval elongated	Fast	Lighter

UV–Vis showed 85.4% DYB decolorization (144 h) by *P. jenningsi*; FTIR confirmed disappearance of azo (–N=N) and amine (C–N) peaks, with appearance of –OH and C=O groups.



GC–MS revealed aromatic amines at day 4, which disappeared by day 6 as long-chain hydrocarbons appeared.

Days	Compound	Abundance (%)	RT (min)	Observation
4th	4-tert-Butylaniline, N-trimethylsilyl	81.13	19.714	Azo bond cleavage of DYB; aromatic amine formation
4th	3-Hydroxyanthranilic Acid, 3TMS derivative	56.20	16.622	DYB nitro reduction & hydroxylation
4th	Bis(2-ethylhexyl) phthalate	33.44	17.082	Complete azo ring cleavage; DYB degradation confirmed
8th	Pentacosane	95.09	8.123	Indicates DYB mineralization
8th	Octacosane, 1-iodo-	95.06	10.032	Indicates DYB mineralization

CONCLUSION

Our findings strengthen the hypothesis that ciliated protozoan, *P. jenningsi* can act as a efficient green biocatalyst for the biodegradation, detoxification, and decolorization of azo dyes.

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