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Antibacterial Effect of Untreated and Treated (Decolorized) Dyes by Agricultural Wastes

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Abstract: Toxicity (antibacterial effect) of untreated and treated (decolorized) dyes on a soil bacterium, *Pseudomonas aeruginosa* was determined. Low cost and easily available two agricultural residues (cotton stalk and apricot seed) as biosorbents were used to remove Astrazone Black and Astrazone Yellow from aqueous solution. The removal of these dyes with agricultural wastes reduced the toxic effect on *P. aeruginosa*. This reduction in toxic effect is important both in respect of environmental biotechnology and waste detoxification. This study showed that these agricultural by-products can be used for decolorization and detoxification of dyes.

Key words: Agricultural wastes, antibacterial effect, astrazone dyes, decolorization, *Pseudomonas aeruginosa*

INTRODUCTION

The removal of dye from textile effluents is one of the most significant environmental problems. Dyes are used in large quantities in many industries including textile, leather, cosmetics, paper, printing, plastic, pharmaceuticals, food, etc. to color their products[1] The extensive use of dyes often poses pollution problems in the form of colored wastewater discharge into environmental water bodies. The presence of very low concentrations of dyes in large water bodies is highly visible and undesirable and also reduces light penetration and photosynthesis. In addition, some dyes are either toxic or mutagenic and carcinogenic[2-5]. Azo dyes are one of the oldest industrially synthesized organic compounds and under anaerobic conditions can be converted to aryl amines which are potentially more toxic than the parent compounds[6,7]. Azo dyes are designed to be recalcitrant under typical product service conditions it is this property, allied with their toxicity to microorganisms that makes biological treatment difficult[8].

Due to low biodegradability of dyes, a conventional biological wastewater treatment process is not very efficient in treating a dye wastewater. The elimination of colored substances in wastewater is based mainly on physical or chemical methods[9]. These methods have several disadvantages like high cost, incomplete removal, low selectivity and high energy consumption. Currently, the most widely used and effective physical method in industry is activated carbon, although running cost are expensive. This mainly due to the chemicals required for regeneration after dye removal[10]. Although, activated carbon removes dyes from solution, they are then present in a more concentrated and toxic form and so their safe disposal increases the cost further. So, effective, more economically alternative and lower cost adsorbents would be of great value.

Among many new technologies, utilizing plant residue as adsorbents for the removal of dyes and metal ions from wastewater is a prominent technology[11]. Various agricultural products and by-products have been investigated to remove dyes from aqueous solutions. These include cotton waste, rice husk, bark[12], peanut hull[2], orange peel[13], sugar cane dust[14], corncob, barley husk[15] and tree fern[16]. The obvious advantage of this method is lower costs involved. New economical, easily available and highly effective adsorbents are still needed.

This study investigated the use of cotton stalk and apricot seed as a biosorbent for color removal of dyes from aqueous solutions. The main purpose of decolorization is detoxification, because dyes in textile wastewater are very recalcitrant and may be inhibitory to microbial consortia of convenential treatment[17]. The aim of the present study was to remove the dyes by agricultural by-product and test the toxic effect of dyes on a soil bacterium *P. aeruginosa* before and after treatment with agricultural wastes.

MATERIALS AND METHODS

Preparation of adsorbents: Two agricultural residue, cotton stalk and apricot seed were used. Cotton stalk and apricot seed were collected from fields in around

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East and Southern East, Turkey. The agricultural wastes were ground and the size distribution was determined by using a sieve.

Preparation of dye solution: The dye stock solutions were prepared by dissolving accurately weighed dyes in distilled water to the concentration of 500 mg L^{-1}. The experimental solutions were obtained by diluting the dye stock solutions in accurate proportions to different initial concentrations.

Decolorization experiments: Decolorization experiments were carried out a rotary shaker at 150 rpm and 30°C using 250 mL flaks containing 50 mL of different concentrations of dye solutions. After shaking the flasks for 30 min., agricultural wastes were separated by centrifugation. The absorbance of the supernatant solution was estimated to determine the residual concentration. Residual dye concentration was determined using absorbance values measured before and after the treatment with spectrophotometer. Results are the mean of at least three replicates.

Toxicity assay: *Pseudomonas aeruginosa* ATCC 10145 which is a soil bacterium was used to test the toxicity of treated and untreated dyes. For this, *P. aeruginosa* was incubated for 18 h at 37°C in nutrient broth medium. Each toxicity test was carried in test tubes in a final volume of 5 ml, containing 100 L bacterial suspensions, 1 mL nutrient broth and the dye sample to be tested[18]. After incubation for 24 h at 37°C, viable cell counts (colony forming units, cfu mL^{-1}) were estimated by plating on nutrient agar plates. Samples of 100 L were taken from the tubes and serial dilutions were prepared with saline. 100 L of the diluted sample was spread onto duplicate nutrient agar plates. The plates were incubated at 37°C for 24 h and then the numbers of viable cell counts were expressed as mean colony forming unit per ml (cfu mL^{-1}).

RESULTS AND DISCUSSION

Effect of initial dye concentration on decolorization: The influence of the initial concentration of dye in solutions on removal of dyes was studied. In our preliminary experiments we have used various adsorbent dose (0.1-0.2-0.5 and 1.0 g) and various adsorbent particle size (25-60-100-270 mesh) for remove to dyes. Results showed that 0.5 g of adsorbent amount had a highest removal capacity and so further experiments were done using 0.5 g adsorbent. Also, most effective adsorbent particle size for cotton stalk and apricot seed were 100 mesh and 270 mesh, respectively and so these meshes were selected for further studies. The experiments were carried out at fixed adsorbent dose (0.5 g 50 mL^{-1}) in the test solution, 30°C temperature, natural pH, fixed agitation (150 rpm) for 30 min. time at different initial concentration of dye (50, 100, 200, 300, 400, 500 mg L^{-1}). The removal of dyes was clearly dependent on the initial dye concentration of the solution (Fig. 1 and 2). Color of Astrazone black was reduced by cotton stalk from 92% to 83% and by apricot seed from 93% to 62% as concentration was increased from 50 to 500 mg L^{-1}. Color of Astrazone yellow was reduced by cotton stalk from 93% to 61% and by apricot seed from 92% to 33% as concentration was increased from 50 to 500 mg L^{-1}.

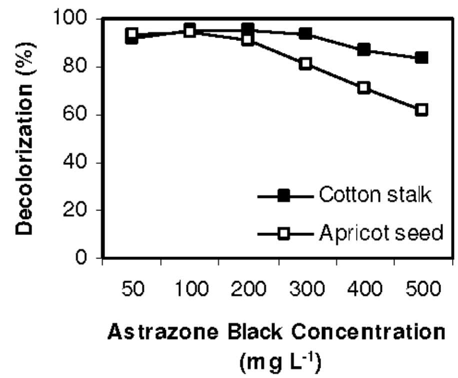

Fig 1. Effect of initial dye concentration on decolorization of Astrazone black by cotton stalk and apricot seed

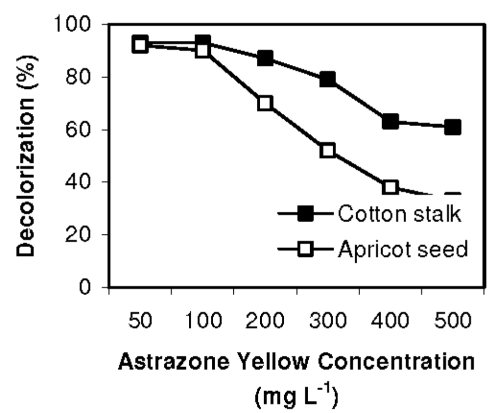

Fig 2. Effect of initial dye concentration on decolorization of Astrazone yellow by cotton stalk and apricot seed

Antibacterial effect of untreated and treated dyes: Dyes can be toxic to many organisms in the aquatic and soil environment. Therefore, the elimination of the dyes in wastewater is an important objective in the search for a method to eliminate its pollution properties.

Antimicrobial effect of the treated dyes on *P. aeruginosa* was tested to evaluate the toxicity after decolorization. In our study it was shown that the dyes used were toxic and their toxic effects increased with an increase in concentration. It appeared that the treated dyes were less toxic. The removal of Astrazone Black and Astrazone Yellow with cotton stalk reduced the toxic effect on *P. aeruginosa* (Table. 1). This reduction in toxic effect is important both in respect of environmental biotechnology and waste detoxification.

Moawad *et al.* (2003) reported that high concentration of azo dyes eliminated microbial colonies due to high frequency of mutation[19]. Apohan and Yesilada (2005) reported that the toxicity of azo dyes could be removed by fungal treatment[17]. Recently, some researchers reported the reduction of toxicity of various dyes after fungal treatment[20,21]. But, there is no information regarding antimicrobial activity of astrazone dyes after decolorization by agricultural wastes. Further toxicity tests are required to support these findings.

Table. 1: The effect of untreated and treated (decolorized) dyes by cotton stalk on the growth of *P. aeruginosa*

	Viable Cell Number (cfu mL^{-1})			
	Astrazone Black		Astrazone Yellow	
Dye (mg L^{-1})	Untreated	Treated	Untreated	Treated
50	1.80×10^{8}	1.72×10^{9}	2.45×10^{8}	1.73×10^{9}
100	2.52×10^{8}	1.57×10^{9}	1.90×10^{8}	1.89×10^{9}
200	1.80×10^{7}	1.36×10^{9}	2.00×10^{8}	2.84×10^{9}
300	1.21×10^{6}	1.03×10^{9}	2.22×10^{6}	2.36×10^{9}
400	0	2.93×10^{8}	0	2.70×10^{9}
500	0	1.84×10^{8}	0	2.01×10^{9}

CONCLUSIONS

The present study showed that the cotton stalk and apricot seed are effective biosorbent at removing dyes from aqueous solution. Therefore, these agricultural materials can be used as cheap and promising biosorbents for dye removal from wastewater of textile industries. Because, most physico-chemical techniques used earlier have several shortcomings which include excess amounts of chemical usage or sludge generation with obvious disposal problems, costly plant requirements or operating expenses[3]. The results obtained in this study for the mechanisms involved in dye removal can be considered as a fundamental step for the representation of the experimental behavior and for development of process design. In addition, decolorization and detoxification ability of the agricultural wastes could be advantageous to integrate decolorization process prior to conventional processes.

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