



## Biotreatment of water polluted with methyl orange dye by using different forms of yeast

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### Abstract

Increasing industrialization and urbanization results in the waste discharge to the environment, which in turn creates more and more pollution. The discharge of toxic effluents from various industries adversely affects the water resources, aquatic organisms and ecosystem integrity. These dyes are carcinogenic for both animal and human beings. Biological treatment either by bacteria, fungi or consortia of both have been reported to reduce the toxicity of dyes to the permissible limit of discharge to the environment. In the present study, the removal of color of methyl orange from aqueous solution had been carried out by Baker's Yeast (*Saccharomyces cerevisiae*). Different factors such as solution pH, dye concentration and biomass dosage at different interval times were experimentally tested using repeated-batch process. The effect of pH on dye bio-removal was investigated at a pH range from 3 to 11. The optimum pH values were 3, 5, 7, 9 and 11 for direct dye removal. The effect of dye concentration was studied using different concentrations of synthetic dye solutions containing 100 – 600 ppm, while the effect of biomass weight was studied in the range 1-5 g/L at pH  $5 \pm 0.1$ . The effect of salinity on dye bio-removal process was investigated at the optimum pH, at concentrations (2, 3 and 4 g/l) of NaCl. The effect of glucose as a nutrient was evaluated for different concentrations of glucose (20, 40 and 60 ml/L).

**Keywords:** Baker's yeast, methyl orange, water treatment.

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### 1. Introduction

The control of water pollution has become of increasing importance in recent years due to increasing in population, development, and assortment of industries [1]. Colored organic compounds that are fully or partially soluble when reached water supplies may cause serious effects to the biota. There are over 100,000 types of dyes available commercially, with a production exceeding one million tons annually [2]. It was estimated that more than 20% of dyes are discharged directly into the environment [3]. So, it is important to remove dyes as compared to colorless compounds as even at low concentrations. Disposal of dyes without pre-treatment into the water streams causes intense ecological disturbance and pollution to marine life [4]. Dyes make up an abundant class of organic compounds characterized by the presence of unsaturated groups (chromophores) such as C=C, N=N and C=N, which are responsible for the dye colours, and of functional groups responsible for their fixation to fibres, for example, NH<sub>2</sub>, OH, COOH and SO<sub>3</sub>H [5]. Although most dyes have a low toxicity, their components and breakdown products can be more toxic. [6]. In addition, improperly discharged industrial dyes pose a hazard to human health. To overcome this problem, much attention has been focused on the effective treatment of dyes discharged from the dyeing industries.

Many methods have been described for color removal from dye-containing wastewater. These methods include adsorption [7], coagulation–flocculation [8], chemical

oxidation (chlorination, ozonation, etc.) [9], electrooxidation [10] or electrocoagulation [11] and photodegradation [5,12]. Existing physical and chemical technologies are expensive and might often produce large amounts of solid wastes [13].

Recently, biological treatment methods using aerobic and anaerobic microorganisms [14,15] have received increasing interest owing to their high effectiveness, lower sludge production and ecofriendly nature [16]. These methods are commonly considered to be the very effective and could be applied for treatment of large scale since they present lower operating costs and promising improved applicability. Many types of microorganisms, such as bacteria [17], fungi [18] and algae [19] were successfully used for decolorization of many types of synthetic dyes in wastewater.

The success of a biological process for color removal from industrial effluents depends mainly on the utilization of microorganisms that effectively destroy synthetic dyes of different chemical structures. Few theses were found in the literature about using yeast for decolorization process [20-21]. In this study, the ability of an important type of yeast *Saccharomyces cerevisiae* (Baker's yeast) to remove methyl orange dye from its aqueous solution, and optimize this process depending on various factors.

### 2. Materials and Methods

Methyl orange dye (4-dimethylaminoazobenzene-4'-sulfonic acid sodium salt), was obtained from Sigma

Aldrich Co., its structures is depicted in Fig. 1. All other chemicals were of analytical reagent grade. Deionized water was used for preparation and dilution of solutions.

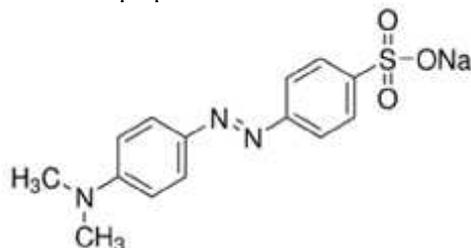


Fig (1): Chemical structure of methyl orange (Mol. wt. 327.34 g/mol and  $\lambda_{\max}$  460 nm)

### 2.1. Microorganism

*Saccharomyces cerevisiae* is commonly known as Baker's and Brewer's yeast [23]. Yeast was the first eukaryote with a complete genome sequence discovered in 1996 [24]. Active dry yeast *Saccharomyces cerevisiae* was purchased from the supermarket.

The purified strain of Baker's yeast (*Saccharomyces cerevisiae*) was used for biodegradation studies. The isolate was identified using the most documented keys in fungal identification [22]. It is a globular shaped unicellular organism with its size ranging between 5-10  $\mu\text{m}$ .

A standard curve for methyl orange was constructed in the range 1-20 mg/L, using a UV/VIS Spectrophotometer–PG instruments Ltd at 465 nm. EUTECH pH-700 pH-meter (Instrument Singapore meter) was used for pH measurements.

A commercially prepared and was kept in a sterile bottle and stored at 4°C. Other chemicals used in the analysis were of analytical grade

### 2.2 Analytical methods

Assays of dye decolorization batch equilibrium studies were conducted in 250 ml conical flasks containing a definite amount of yeast with known initial dye concentration. The pH of the solution was adjusted to the required value and the experiments were conducted at fixed temperature. At different time intervals, about 3 ml of the dye solution were withdrawn and centrifuged to obtain a clear solution. The equilibrium dye concentration was measured by using the spectrophotometer at  $\lambda_{\max}$ .

### 3. Results and discussion

The degradation of the selected azo dye (methyl orange) was assessed in conjunction with strain of Baker's yeast which is particularly widely used in industrial operations. The yeast strain possessed the most pronounced ability to remove the dye from aqueous solution [25]. The decrease of color biodegradation in the contact solution with time is studied in all tests at  $37 \pm 2$  °C and at a stirring rate of 180 rpm then we measured the decolorization of colour .

#### 3.1. Effect of pH

The pH is one of the most important factors controlling the biodegradation of a dye. The effect of solution buffering on methyl orange removal was examined in a batch equilibrium system at a pH values of 3.0, 5.0, 7.0, 9.0 and 11.0 as shown in Fig. 1 (a and b). Batch experiments were carried out at a dye concentration of 50 ppm (50 ml) for 48 h. The results indicated a good dye removal at pH 5.

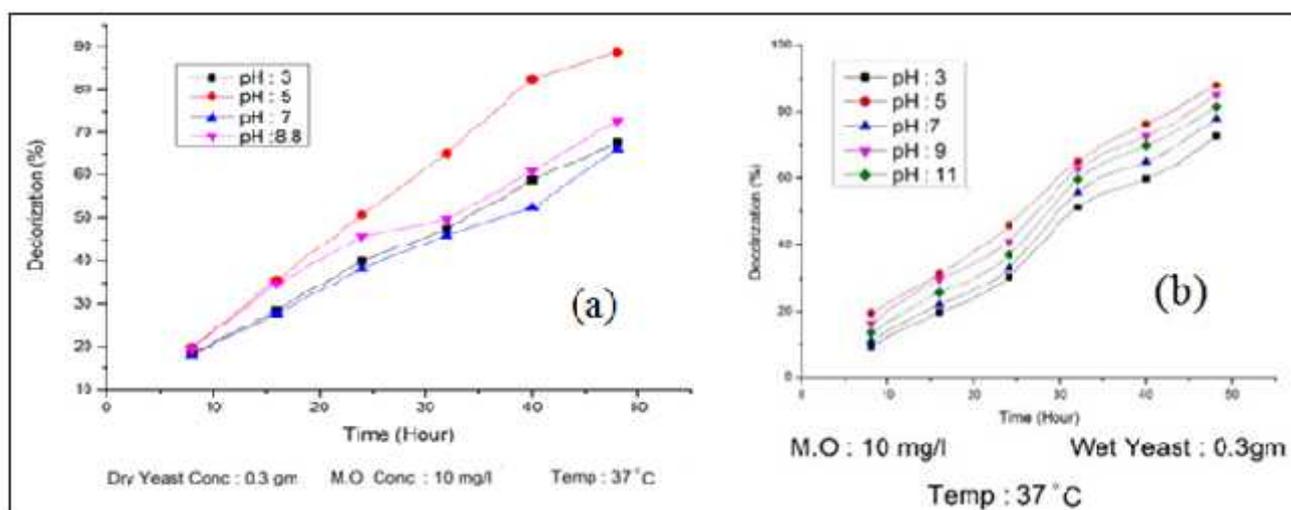


Fig (2): Decolorization of M.O. by (a) Dry and (b) wet yeast at different pH values.

### 3.2. Effect of different yeast doses

The effect of different doses of yeast (0.1–0.5 g/L) on methyl orange at pH 5.0 on decolorization of the dye was studied for 48 h. The selection of this pH was based on the optimal pH study. It was observed that the increase in yeast dose (dry or wet) caused a corresponding increase in decolorization efficiency. It was found that for the two

types of yeast, increasing the yeast dose from 1 to 5 g/L leads to a corresponding M.O. dye removal from 63.2 to 90.7% for dry yeast and from 63.5 to 94.5% for wet yeast, after 48 h (Fig 3). This behavior is expected, as the increase of yeast increases the efficiency of the dye biodegradation.

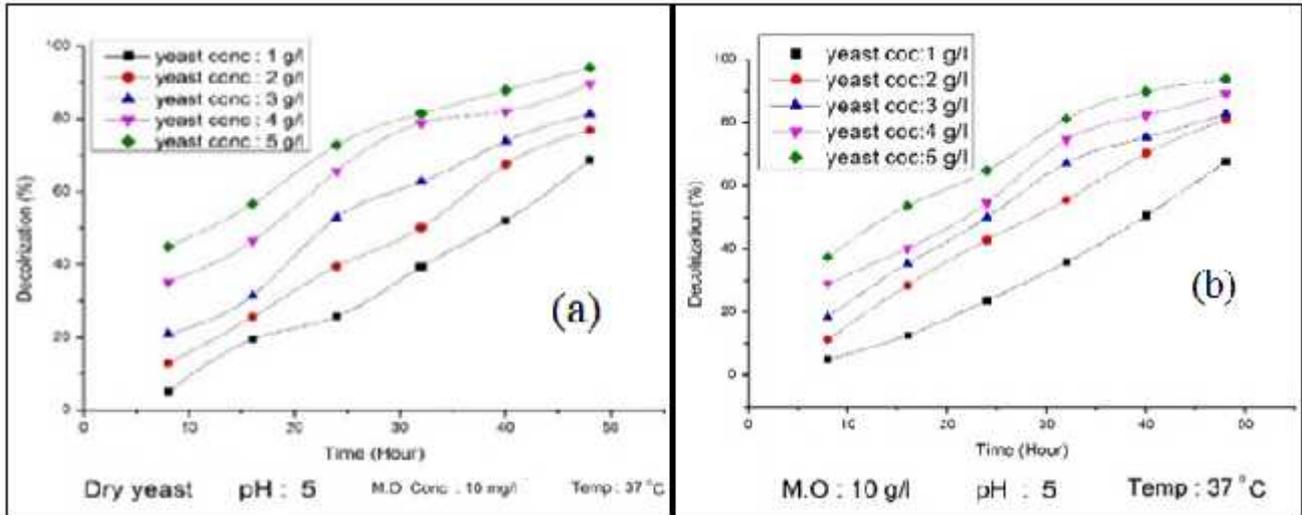


Fig (3): Decolorization of M.O. by different doses of (a) dry and (b) wet yeast.

**3.3. Effect of salt concentrations**

The effect of different salt concentrations (2.0, 3.0 and 4.0 g/L) on M.O. decolorization at pH 5.0 was studied for 48 h. The selection of this pH was based on the optimal pH determined for pH study. For both types of yeast, it was found that increase of salt content caused an increase in dye removal percent. It was found that increase of salt (NaCl) affects the efficiency positively. High salt levels can dehydrate anaerobic yeast cells because of osmotic pressure. Anaerobic digester was much more sensitive to chlorides than activated sludge [26].

Baere et al. (1984) [27] examined influence of high NaCl on methanogenic activity on the percentage of dye

removal increases with the increasing the initial salt (NaCl) concentration; this may be due to the saturation of the sorption sites on the biosorbent as the concentration of the dye increases. The actual amount of dye adsorbed increases with increasing initial yeast concentration, i.e., the increase in adsorption is limited with biosorbent dose. This was confirmed by other investigators and they attributed this phenomenon to the increase in the driving force of the concentration gradient, with the increase of the initial yeast concentration. Hence a higher initial concentration of the yeast will enhance the adsorption process [26,27].

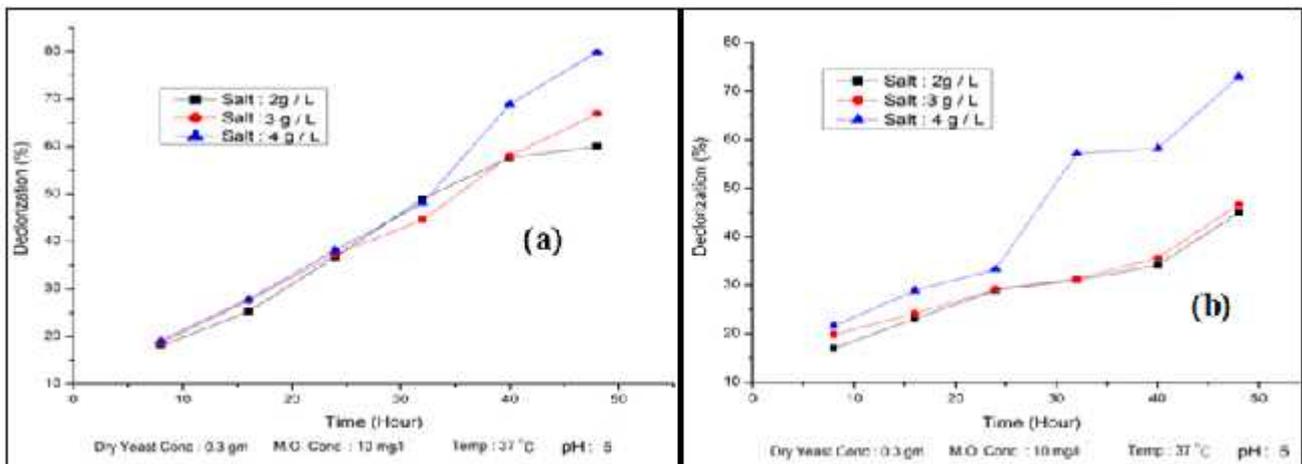


Fig (4): Decolorization of M.O. by yeast for different salt concentration; (a) dry and (b) wet yeast.

**3.5. Effect of glucose concentrations**

For many microorganisms, including yeasts, glucose is the preferred carbon and energy source. Glucose affects many important traits of the yeast: growth rate, fermentation capacity and stress resistance. More specifically, yeast cells have developed mechanisms to respond to extreme variations in nutrient availability by modulating their growth and metabolism. Evidently, in the presence of all these sugars, glucose has the fastest rate of carbon dioxide production, followed by fructose, maltose and sucrose [28].

However, these processes will depend on the presence or absence of oxygen. In aerobic conditions, pyruvate will breakdown into carbon dioxide and water to produce adenosine triphosphate (ATP) energy. This is known as oxidative phosphorylation [29]. In anaerobic conditions, the process observed is fermentation, the breakdown of sugars into carbon dioxide and ethanol known as the Gay-Lussac equation [30]. This can be observed in the following equation:



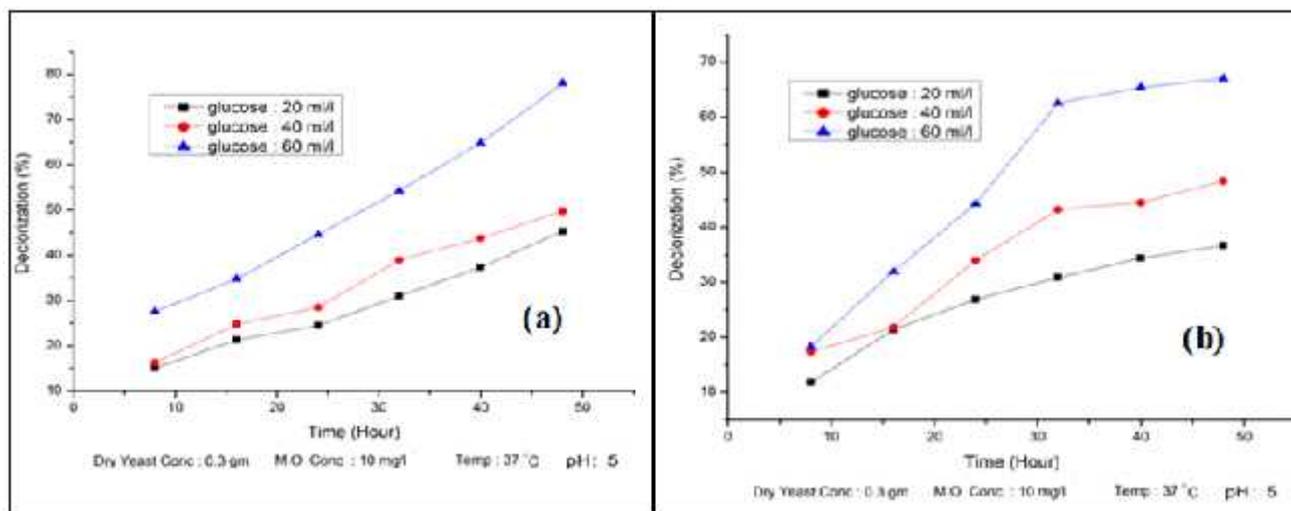


Fig (5): Decolorization of M.O by yeast for different glucose concentration; (a) dry and (b) wet yeast.

### Conclusion

According to previous experiments and the results obtained from changes the factors effects on yeast *saccharomyces cerevisiae* efficiency for dye waste degradation we conclude that firstly: The ability of *saccharomyces cerevisiae* to remove color of M.O from solutions increased with increasing its concentration in the solution, hence increasing its efficiency. Secondly: To

obtain the best conditions for achieving the highest remove of dye color from the solution we used different concentrations of the pH solutions we found that the best efficiency of *saccharomyces cerevisiae* at pH:5 and so on the best efficiency of it at salinity concentration at 4.0gm/L and glucose concentration at 60 ml/L.

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