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Flocculation Of Saccharomyces Cerevisiae With Grewiasppbiopolymers

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ABSTRACT:

Removal of Yeasts *(Saccharomyces cerevisiae*) by coagulation/flocculation with natural extracted polymer from *Grewiaspp* plant originated from Maroua (Cameroon) was studied. Experiments were conducted in a jar-test at different pH [4, 7.9], ionic strength [10^{-3}, 10^{-2}, 10^{-1} (M)], concentration of coagulant / flocculant (mg / L) [0.125-0. 25-0.375-0.5-0.625], turbidity [60NTU], and stirring speed / time [$550s^{-1}$/5 min and $100s^{-1}$/15 min]. Results showed that maximum reduction rate (35%) was obtained forpH 4, ionic strength 10^{-1}M, and polymer concentration of 0.375 mg / L. With $CaCl_2$, removal rateof yeast improves up to 90%. Charge neutralization and adsorption at pH 7, adsorption and bridging at pH 4, and flocculation at pH 9 were identified as mechanisms involved in this process.

INTRODUCTION

Brewing processneeds an average of $10m^3$of water per m^3 of beer. This large amount of water is used for cooking, cooling, heating or washing [1], and generates significant amounts of effluent. Waste water resulting from washing operations contains organic matter, highly enrich of yeast(*Saccharomyces cerevisiae) when* it comes from the fermentation stage[2]. Those effluents of high DBO5 may contribute to environmental pollution if rejected without any treatment.They can be treated by several treatments including biological, physical and chemical ones. Most of the time decantation process is used. Unfortunately, as yeast is of small size and difficult to settle easily, natural decantation is time consuming and there's a need to increase their settling time, and the separation efficiency. This can be done by coagulation / flocculation[3].

Coagulantscan be synthetic or natural and of various origins. Syntheticcoagulants are very effective in removing particulate matter and are widely used in water and wastewater treatment. They can be of cationic, anionic or nonionic type[4]. The most commonly used are aluminum or iron salts, polymerized forms and polyacrylamide[4]. The use of aluminum salts generates large non-biodegradable quantities of toxic sludge and has been reported to be important in Alzheimer diseases[5]. The high cost of synthetic polyelectrolytes, the dispersion of toxic acrylamide monomers especially for aquatic organisms (algae, fish, daphne ...)[4], an alternative was considered therefore to overcome the drawbacks series of environmental, economic and health posed by synthetic coagulants. Several studies have been conducted on natural polymers that are available, inexpensive and biodegradable. The extracts of several plants of different families

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have been tested for this purpose conclusively in coagulation / flocculation water. Plants like*Moringaoleifera, Strychnospotatorum, JatrophacurcasH sabdariffa, Cangustifolia*[6], *Phaseolus vulgaris*[7] are commonly used. In Cameroon, *Grewiaspp*is widespread and used by small and medium enterprises, local industries and the population in the clarification of water wells and a local beer (bilibili).

The objective of this study is to evaluate the efficiency of aqueous extracts of*Grewiaspp* as a coagulant / flocculant in the clarification of a water-rich yeast *Saccharomyces cerevisiae* type.

I- MATERIALS AND METHODS

A. Extraction of coagulant / flocculant

*Grewiaspp*barks were originated from Maroua, a town in the far north region of Cameroon. After drying at 45°C for 48 h, crushing and sieving, the resulted powder fraction used hada diameter between 160 µm and 250 µm. Polymer extraction was performed in a batch mode using the central composite design after selection of significant parameters with the Plackett-Burman plan. For polymer extraction, 1.5 g of powder suspended was introduced in 100 mL of distilled water at a fixed pH (8), ionic strength: (0.022 M), stirring speed and time ($250s^{-1}$ for 19 min), and temperature(43 °C) for 1 H. The resulting mixture was separated by centrifugation (4000 s^{-1} for 39 min) and the supernatant which represents the active extract (coagulant) was kept at 4 ° C after the addition of atrazide 10 ppm to avoid bacterial growth.

B. *Saccharomyces cerevisiae*

Lyophilised*Saccharomyces cerevisiae* was introduced into physiological solution containing chloramphenicol for 24 hours, and the yeast concentration of this initial suspension was evaluated using dilution technique on plate count agar (PCA) medium with chloramphenicol, and incubation at 27 °C. After two days of incubation yeasts were removed from the culture medium and washed with distilled water using centrifugation at 5000trs/min for 15 min at 5 °C [6]. Resulting pellets containing live yeast were stored in physiological solution at 4 °C.

Zeta potential of *Grewia* spp. particles was carried out using a Zeta sizer2000 and the S. Cerevisae particle size was determine using a Master sizer 2000. Previous studies on this plant have reported the presence of polysaccharides as main polymers extracted from this plant[8].

C. Coagulation tests

Experiments were conductedin a jar test (Fisher-Bioblock) with a synthetic effluent containing yeastat a fixed turbidityof 60 NTU. 800 mL of suspension was coagulated at various concentrations (0.125, 0.25, 0.375, 0.5, 0.625 mg / L) under a shear rate of 550 s^{-1} for 5 min.After addition of *Grewia* spp.biopolymers the shear rate was reduced to 100 s^{-1} for 15 min and the suspension allowed to settlefor 30 mn. The residual turbidity was monitored at intervals of 10 min and the coagulant activity was determined using the following formula:

$$\text{Coagulant activity (\%)} = \frac{T_i - T_f}{T_i} \times 100$$ where Ti is the initial turbidity and T_f the final one

RESULTS AND DISCUSSION

Results of the coagulant activity of SC with *Grewia*spp. concentration at different pH and ionic strength are presented on figure 1(a) for pH

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4, 1(b) for pH 7 and 1(c) for pH 9. As shown on figure 1(a), increasing the biopolymer concentration up to 0.3 mg/L results in a slight increase of the coagulant activity from 15 to 20% for 10^{-3} M KCl and from 15 to 35 % for 10^{-1} M KCl, while it remains quite constant at 10^{-2} M KCl. At 0.4 mg/L of biopolymer concentration, a drop of almost 5% on the coagulant activity is observed at 10^{-2} and 10^{-3} M KCl, while an increase is found at 10^{-1} M KCl. Beyond 0.4 mg/L constant coagulation activity is found at 10^{-1} MKCl (32%) and 10^{-3} M KCl (22%).

On figure 1(b), the coagulant activity varies from 18% to 28%. The minimum value is obtained with 0.1 mg/L of biopolymer at 10^{-1} M KCl, and the maximum is also found with 10^{-1}M KCl, but at a polymer concentration of 0.4 mg/L. up to 0.3 mg/L slight difference on coagulant activity is observed between 10^{-3} and 10^{-2}M KCl, and it is observed a drop at 0.1 mg/L of polymer and a slight increase up to 0.3 mg/L. After 0.4 mg/L almost constant values of coagulant activity is observed, whatever the ionic strength.

On figure 1(c), the coagulant activity varies from 6% to 18%. The minimum value is obtained with 0.1 mg/L of biopolymer at 10^{-1} M KCl, and the maximum is found with 10^{-3}M KCl at a polymer concentration of 0.5 mg/L. Up to 0.5 mg/L coagulant activity increases and is almost the same observed for 10^{-3} and 10^{-2}M KCl, but with 10^{-1} M KCl it remains much ore lower.The effect of the ionic strength is clearly exhibit when at pH 4. At this pH, coagulation activities at 10^{-3}M and 10^{-1}MKCl, can be explained by an adsorption of yeast on biopolymers. As the surface charge of *Grewia*spp. Extracted biopolymers Is close to 0 mV at pH 4 and that of yeast is negatively charge, it is obvious that attraction is much more important with the ionic strength. Also at pH 4 the biopolymers configuration is different according to the rheology of those suspensions and particle stress

The presence of salts promote the salting out effect [9].Which contributes to render insoluble the polymer and thereby enhance coagulation or flocculation. At pH 7 and at pH 4, aggregation of yeast is attribute to the compression their electrochemical double layer as it'sincreases with the amount of salt. In these conditions, the flocculation could be explained bymolecular interaction established between proteins and carbohydrates of the cell surface and receptors of another cell [10].After flocculation, the adsorbed polymer on yeast has sufficient particle size and density to settle. This could be explained by bridging mechanisms; on the one hand by the negative charges carried by the yeast and the polymer and on the other hand by the presence of potassium salts that result in intracellular calcium efflux probably involved in the bridging of the polymer and yeasts [11]. However, with increasing pH (from 4 to 9) a decrease in the abatement efficiency is observed, and may be a result of the change in conformation of the polymer with pH and or salt concentration. For ionic strengths of 10^{-3}M, 10^{-2}M, 10^{-1}M, it is observed a coagulation / flocculation due to the effects of salting in and out, the staff turnover rate remained low.At pH 4 and 10^{-1} M of KCl, the polymer has a linear conformation that enhances flocculation [4] up to 35%. Increasing the pH up to a neutral value would favour intramolecular bonds, resulting on the change from a linear configuration to a globular one, resulting in the vacancy and the inaccessibility of some adsorption sites, an thus less particle aggregation and settling.

Figure 2 presents the influence of $CaCl_2$ coagulation / flocculation has indeed confirmed

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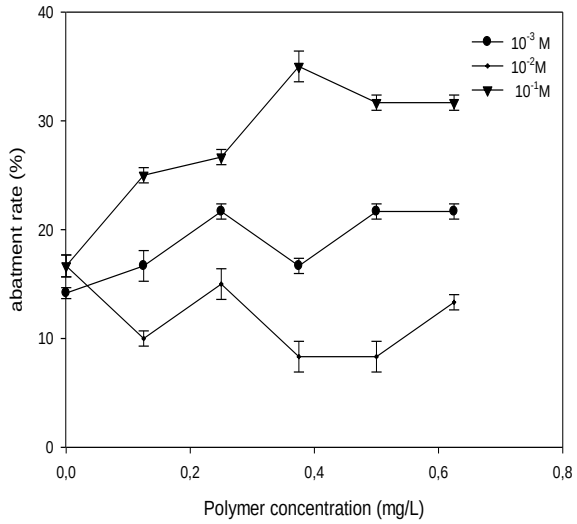
the above explanation. Indeed in the presence of $CaCl_2$ turnover rate is 90%. This shows that calcium chloride promotes aggregation of large flocs and thus improves the settling velocity of flocs, which results in increase rates of depression.

This figure highlights the fact that indeed the biopolymer is able to form yeast aggregates, the resulting flocs do not settle easily. By cons in the presence of $CaCl_2$, decanting is better, indicating a character of weight calcium chloride.



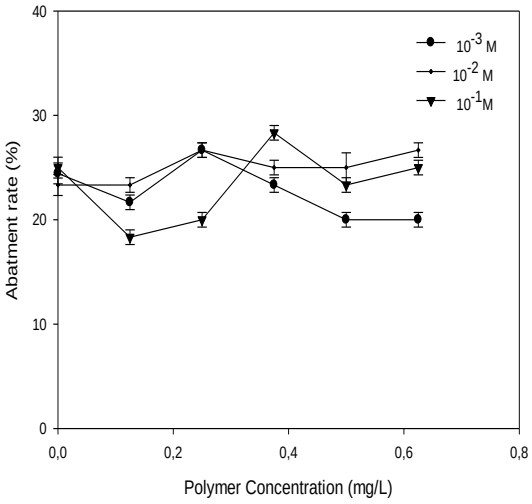
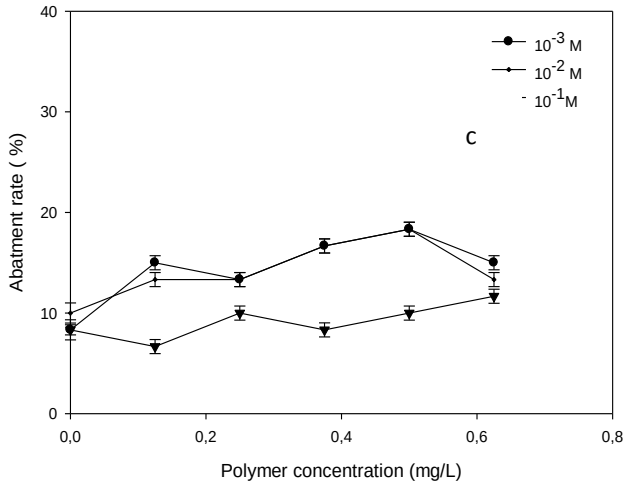


Figure 2: abatement rate versus pH



Figures 1: abatement rate versus polymer concentration at different ionic strength:pH 4 (a) 7 (b) 9 (c)

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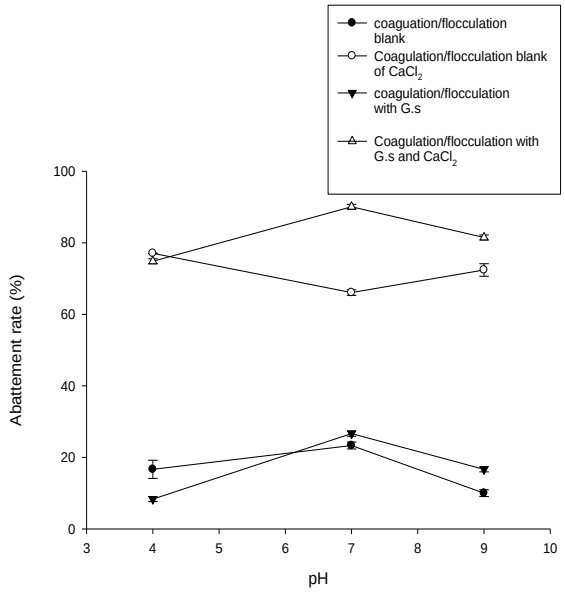

Figure2: abatement rate versus pH

IV- CONCLUSION

The objective of this work was to evaluate the efficiency of clarification of *Saccharomyces cerevisiae suspensionsusing biopolymers of Grewia*sppas a coagulant. It comes out that the pH, ionic strength and polymer concentration has a great influence on the overall turbidity removal. The maximum reduction rate of 35% was found for the following conditions: pH 4, ionic strength: 10^{-1}M, polymer concentration: 0.375 mg / L, and charge neutralization and adsorption at pH 7, adsorption and bridging at pH 4, and flocculation at pH 9 were the main mechanisms observed.

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