

Spent Brewer's Yeast as Biosorbent for Some Heavy Metal Ions

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Abstract

Yeasts are efficient biosorbents for heavy metal ions. The purpose of this experiment was to demonstrate the involvement of the free amino acids present in Spent Brewer's Yeast (BSY) in the coordination of copper ions of Cu²⁺, Pb²⁺, Zn²⁺ ions. Amino acids involvement in metal biosorption could be attributed to their capacity to coordinate metal ions. As biosorbents was used spent brewer's yeast type *Saccharomyces cerevisiae* purchased from the local brewery. The metal ion solutions were prepared in concentrations of 1 mg/L using their salts. The experiments were conducted at pH=5, which is the most appropriate for amino acids and metal ions to avoid precipitation. The amino acids were quantified by HPLC-DAD/-ESI-MS chromatography method. There were 24 amino acids quantified by HPLC method in brewer's yeast and their profile differs after biosorption. The significant lower concentrations of amino acids remaining after biosorption can be explained by their involvement in the complexation of metal ions.

Keywords: Brewer yeast; Amino acids; Biosorption; Heavy metals

Introduction

Biosorption can be defined as a process that uses microorganisms to capture metal ions from solutions by microbial cells. This process can be successfully used to immobilize metal ions in waste waters [1,2]. The sequestering refers to physicochemical mechanisms of inactive metal uptake by microbial biomass [3]. Metal ions can be immobilized by complexation, coordination, ion exchange, precipitation, reduction by some parts of the cell [4,5]. Inactive microbial cells can only immobilize metals by biosorption, whereas active microbial cells may immobilize soluble metal species both by biosorption and by other mechanisms due to the microbial metabolism [6-8]. A significant capacity for metal ions binding possess some functional groups present in amino acids, proteins or polysaccharides [9,10]. Brewer's yeast is an ingredient used in the production of beer and it is made from one celled fungus, Saccharomyces cerevisiae. The live cells and non living cells of yeast biomass have proved to be efficient for removal of Ag, Au, Cd, Co, Cr, Cu, Ni, Pb, Zn from aqueous solution. Literature studies demonstrated that S. cerevisiae can remove toxic metals, recover precious metals from aqueous solutions to various extents [11]. The purpose of this experiment was to demonstrate the involvement of the free amino acids present in spent brewer's yeast in the coordination of copper ions of Cu²⁺, Pb²⁺, Zn²⁺ ions as it is known that amino and carboxyl groups could be responsible for complexation.

Materials and Methods

Saccharomyces cerevisae biomass was supplied as a by-product from a local brewery. The biomass was stirring followed by centrifugation at 3000 rpm for 20 min in order to remove other particles. Metals solution were prepared using the mixture of their salts CuSO4•5H₂O, PbNO3•2H₂O, ZnSO₄ of analytical reagent grade. The metal ion solutions were obtained by dissolving their salts in deionized water, and the concentrations obtained were 1 mg/L. The experiment was performed by mixing 50 ml solution spent brewer's yeast with 50 ml mixture of metals ions-containing solution in 250 ml Erlenmeyer flask on an orbital rotary shaker at 120 rpm for 120 min. The experiment was conducted at pH=5 and was established by adjusting it with HCl 0.1M or NaOH 0.1M solutions. For quantification of the free amino acids from BSY before and after biosorption by HPLC method, the samples were centrifugated at 2500 rpm for 15 min and the supernatant was analyzed. The HPLC analysis was performed on an Agilent 1200 system equipped with a binary pump delivery system LC-20 AT (Prominence), a degasser DGU-20 A3 (Prominence), diode array SPD-M20 A, UV-VIS detector (DAD). Amino acids (100 μl) from control brewer's yeast and samples provided

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Table 1: Amino acids in control BSY and BSY after biosorption (expressed in nmoli/ml).

| | Amino acid | Control BSY | BSY after Biosorption |
|----|-------------------------|------------------|-----------------------|
| 1 | Arginine | 7188.95 ± 25.94 | 1298.25 ± 4.49 |
| 2 | Citruline | 787.34 ± 2.73 | 135.80 ± 0.57 |
| 3 | Glutamine | 5015.70 ± 23.07 | 618.42 ± 1.39 |
| 4 | Serine | 10556.71 ± 48.78 | 1190.01 ± 3.59 |
| 5 | Anserine | 6048.53 ± 26.39 | 1075.52 ± 3.37 |
| 6 | 4-Hydroxyproline | 9810.13 ± | 1220.72 ± 4.09 |
| 7 | Glycine | 10223.28 ± 44.45 | 1787.63 ± 7.61 |
| 8 | Threonine | 5715.06 ± 23.57 | 614.11± 2.07 |
| 9 | Alanine | 21336.87 ± 86.68 | 3984.77 ± 16.93 |
| 10 | β-Amino isobutyric acid | 8369.91 ± 37.84 | 1079.49 ± 3.38 |
| 11 | α-Amino butyric acid | 557.37 ± 1.78 | 360.73 ± 0.96 |
| 12 | Ornithine | 707.21 ± 2.63 | 81.65 ± 0.33 |
| 13 | Methionine | 2465.57 ± 10.32 | 467.29 ± 1.27 |
| 14 | Proline | 11785.82 ± 49.34 | 1245.59 ± 4.22 |
| 15 | Aspartic acid | 5337.96 ± 17.68 | 176.27 ± 0.58 |
| 16 | Lysine | 3636.88 ± 14.18 | 808.00 ± 2.83 |
| 17 | Histidine | 2565.40 ± 9.82 | 488.37 ± 1.26 |
| 18 | Valine | 11343.37 ± 47.15 | 2894.18 ± 11.47 |
| 19 | Glutamic acid | 19157.83 ± 76.98 | 355.77 ± 1.05 |
| 20 | Tryptophan | 1737.76 ± 7.14 | 328.12 ± 0.84 |
| 21 | Leucine | 6986.08 ± 28.93 | 1523.29 ± |
| 22 | Phenylalanine | 3504.44 ± 15.12 | 963.46 ± 4.68 |
| 23 | Isoleucine | 8043.10 ± 33.72 | 1443.23 ± 5.16 |
| 24 | Tyrosine | 2520.72 ± 8.31 | 666.98 ± 2.23 |

after biosorption of metal ions were identified using an EZ: Faast Kit for free amino acids, provided by Phenomenex (USA).

Results and Discussion

The main amino acids quantified by HPLC method were presented in Table1. Amino acid analysis was performed on the control yeast and on the samples resulting after metal ions biosorption. The concentrations of amino acids are different between control brewer's yeast and samples resulted after biosorption of metal ions due, probably, to the complexation of copper, zinc and lead. It can be assumed that the free amino acids in yeast, due to their functional groups, have the ability to complex these metals, and the obtained compounds have different retention times and peaks area than free amino acids. The most abundant amino acids in BSY were Alanine, Glutamic acid, Proline, Serine, Glycine. The biosorption capacity varies from Glutamic acid with 1.85% concentration remained in

solution to α -Amino butyric acid with 64.63%. Glutamic and Aspartic acid provided the best metal ions biosorption capacity at pH=5. The decrease of the amino acid concentrations in the samples incubated with metal ions solution may be due to their coordination with the amino and carboxyl groups and to the formation of complex metalligand amino acid. Another explanation regarding the changes in amino acids profiles and content is that it could be affected by autolysis and fermentation conditions like time, temperature, pH [12].

Conclusion

This study intends to find a possible explanation for the metal ions biosorption by free amino acids of brewer's yeast. The preliminary results lead to the conclusion that the constituent amino acids of yeast have the ability to bind copper, zinc, lead ions from their solutions. This process is dependent on experimental conditions like pH, time of biosorption, temperature, biomass dose and of course, further investigations are required.

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