Soybean Lecithin Based Reverse Micellar Extraction of Pectinase from Synthetic Solution

Authors: Sivananth Murugesan, I. Regupathi, B. Vishwas Prabhu, Ankit Devatwal, Vishnu Sivan Pillai

Abstract: Pectinase is an important enzyme which has a wide range of applications including textile processing and bioscouring of cotton fibers, coffee and tea fermentation, purification of plant viruses, oil extraction etc. Selective separation and purification of pectinase from fermentation broth and recover the enzyme form process stream for reuse are cost consuming process in most of the enzyme based industries. It is difficult to identify a suitable medium to enhance enzyme activity and retain its enzyme characteristics during such processes. The cost effective, selective separation of enzymes through the modified Liquid-liquid extraction is of current research interest worldwide. Reverse micellar extraction, globally acclaimed Liquid-liquid extraction technique is well known for its separation and purification of solutes from the feed which offers higher solute specificity and partitioning, ease of operation and recycling of extractants used. Surfactant concentrations above critical micelle concentration to an apolar solvent form micelles and addition of micellar phase to water in turn forms reverse micelles or water-in-oil emulsions. Since, electrostatic interaction plays a major role in the separation/purification of solutes using reverse micelles. These interaction parameters can be altered with the change in pH, addition of cosolvent, surfactant and electrolyte and non-electrolyte. Even though many chemical based commercial surfactant had been utilized for this purpose, the biosurfactants are more suitable for the purification of enzymes which are used in food application. The present work focused on the partitioning of pectinase from the synthetic aqueous solution within the reverse micelle phase formed by a biosurfactant, Soybean Lecithin dissolved in chloroform. The critical micelle concentration of soybean lecithin/chloroform solution was identified through refractive index and density measurements. Effect of surfactant concentrations above and below the critical micelle concentration was considered to study its effect on enzyme activity, enzyme partitioning within the reverse micelle phase. The effect of pH and electrolyte salts on the partitioning behavior was studied by varying the system pH and concentration of different salts during forward and back extraction steps. It was observed that lower concentrations of soybean lecithin enhanced the enzyme activity within the water core of the reverse micelle with maximizing extraction efficiency. The maximum yield of pectinase of 85% with a partitioning coefficient of 5.7 was achieved at 4.8 pH during forward extraction and 88% yield with a partitioning coefficient of 7.1 was observed during backward extraction at a pH value of 5.0. However, addition of salt decreased the enzyme activity and especially at higher salt concentrations enzyme activity declined drastically during both forward and back extraction steps. The results proved that reverse micelles formed by Soybean Lecithin and chloroform may be used for the extraction of pectinase from aqueous solution. Further, the reverse micelles can be considered as nanoreactors to enhance enzyme activity and maximum utilization of substrate at optimized conditions, which are paving a way to process intensification and scale-down.

Keywords: pectinase, reverse micelles, soybean lecithin, selective partitioning

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