"Study of Lipase Partitioning Behaviour in Ionic Liquid-based Aqueous Two-phase Systems to Improve Bio-separation Process of Microbial Lipase from Fermentation Broth"

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In view of the awareness of the deteriorating environment due to the use of volatile organic solvents, the research focus is directed to develop alternative environmentally friendly solvents. Within this context, ionic liquids (ILs) have emerged as viable replacements for traditional organic solvents due to their negligible volatility, non-flammability and high chemical/thermal stability. Moreover, one of the major attractions of ILs is their "designer solvent" properties. The characteristics of ILs can be tailored made by appropriate selection of their cations and anions. This advantage allows the development of an IL for a specific task.

The present work presents the generation of novel ILs possessing buffering capacity and high biocompatibility, aiming for their applicability in bioprocessing technology. The widely used biological buffers, more specifically, Good's buffers (GBs) that introduced by Norman Good et al. in 1966, were used to prepare the anions of ILs. The GBs, namely 2-hydroxy-3-morpholinopropanesulfonic acid (MOPSO), N-[tris-(hydroxymethyl)methyl]-3-amino-2-hydroxypropanesulfonic acid (TAPSO), 2-[bis(2hydroxyethyl)amino]ethanesulfonic acid (BES) and 3-(cyclohexylamino)-2-hydroxy-1propanesulfonic acid (CAPSO) were coupled with tetrabutylammonium, tetrabutylphosphonium and cholinium cations via an acid-base neutralization reaction to produce 12 ILs. These ILs were designated as Good's buffer ionic liquids (GBILs).

The GBILs were characterized for their structures, thermophysical properties, buffering capacity, enzyme-compatibility and ecotoxicity. Furthermore, the feasibility of these GBILs to act as phase-forming components in aqueous biphasic systems (ABS), a type of liquid-liquid extraction technique, was investigated. Additionally, the potential of GBIL-based ABS were studied by evaluating the partition behaviour of lipase in these systems and the possible interactions between GBIL ions and the lipase residues. After selecting the suitable GBIL-based ABS, the systems were investigated for their applicability to recover and purify lipase produced by the submerged fermentation of *Burkholderia cepacia* ST8. Besides, the aqueous solutions of GBIL buffer were applied as recovery solvents in the ultrasonication-assisted extraction of proteins from *Chlorella vulgaris*. The results reported here open a promising prospect for the usefulness of the GBILs as extraction solvents in bioprocessing technology.

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