

## Alternative protein purification technologies using immobilized ionic liquids as chromatographic ligands

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**Abstract:** The increase of life expectancy has been accompanied by an increment of chronic diseases, with biopharmaceuticals becoming one of the most effective clinical treatments for a broad range of diseases. However, their manufacturing can be time-consuming and expensive, thus limiting their utilization. Their manufacturing is typically divided in the upstream and downstream phases with the latter being responsible for most of the production costs of biopharmaceuticals (50–90%), mostly due to chromatographic processes. Chromatography is usually considered the gold-standard of purification techniques, allowing the separation, and purification of compounds present in a complex mixture based on different properties including size and shape, charge, or hydrophobic groups present of the surface [1]. Nevertheless, current extraction, separation and purification processes are still trying to overcome some drawbacks, especially when it comes to affinity chromatography due to ligands poor selectivity and specificity as well as their associated cost. To overcome these limitations, recent developments on extraction and purification processes using “greener” and more sustainable materials such as the use of supported Ionic Liquids (SILs). One of the main advantages of the use of SILs is that the *designer solvent* character of ILs is transferable to SILs through the immobilization of ILs in suitable support materials allowing the choice of more biocompatible and less expensive materials for chromatographic matrixes such as silica. These materials have been successfully applied for the recovery of nucleic acid-based biopharmaceuticals [2,3] however their utilization for recombinant protein recovery has not yet been investigated. In this work, several SILs were applied as chromatographic stationary phases for IFN $\alpha$ -2b purification with different elution conditions being applied to increase the purity of the target protein, by exploring different types of interactions. In general, the SIL modified with the imidazolium-based ionic liquid demonstrated the highest ability to purify IFN $\alpha$ -2b purification, either in conditions favoring electrostatic interactions or hydrophobic interactions. Overall, this work demonstrates that ILs as ligands in solid supports allow the establishment of a multitude of different molecular

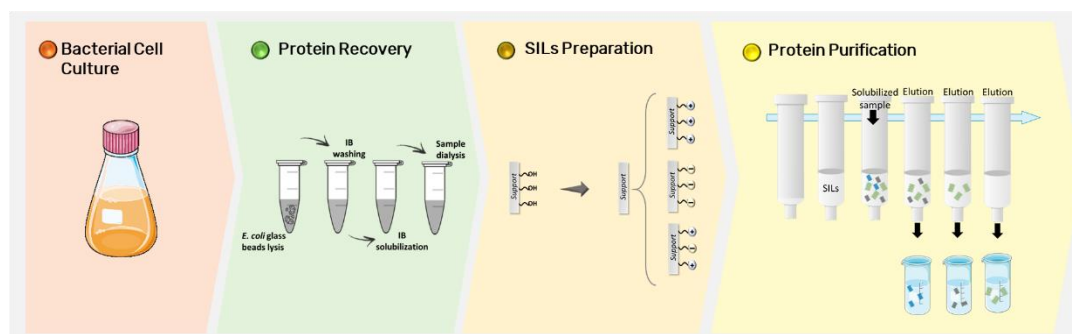
interactions, which can be explored to enhance the purification of recombinant therapeutic proteins toward more sustainable processes.

#### References:

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**Schematic representation of the purification process of IFN $\alpha$ -2b using supported ionic liquids.**