Stabilization and purification of RNA using biobased ionic liquids

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Abstract:

The ubiquitous unstable nature of RNA has been preventing its widespread use in the development of novel therapies. Moreover, the traditional RNA extraction and purification techniques are laborious and require highly toxic reagents, demanding for improved technologies able to provide RNA with high purity and biological activity.

To overcome the described bottlenecks, ionic-liquid-based aqueous biphasic systems (IL-based ABS), which are mainly composed of water, are herein investigated as more sustainable and efficient techniques for RNA purification. Based on the well-known high affinity between amino-acids and RNA and the favourable nucleic acids-stabilization properties exhibited by amino-acid-based ILs (AA-ILs), they were selected as components of ABS formulations. After identifying the most promising systems able to protect RNA from ribonuclease-mediated degradation, the ultimate goal of this work is to purify RNA from a complex recombinant lysate, envisaging the development of integrated purification-preservation platforms.

AA-ILs comprising L-arginine, L-tryptophan and L-lysine as cations combined with chloride or DL-aspartate ([Arg]Cl, [Trp]Cl, [Lys]Cl, [Arg][Asp], [Trp][Asp], and [Lys][Asp]) were synthesized and characterized, and their ability to form two phases with distinct salts and polymers investigated. With exception of [Trp]Cl and [Lys][Asp], all the remaining AA-ILs form ABS with polypropylene glycol 400 (PPG 400) and were further selected for initial studies on the stability and integrity of RNA to assess its potential to act as RNA preservation media. Several promising IL-based ABS were found and are

currently under investigation as integrated purification-preservation platforms for RNA envisaging its use as biotherapeutics.

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