

Hexafluoroisopropanol-based Deep Eutectic Solvents for High-performance DNA Extraction

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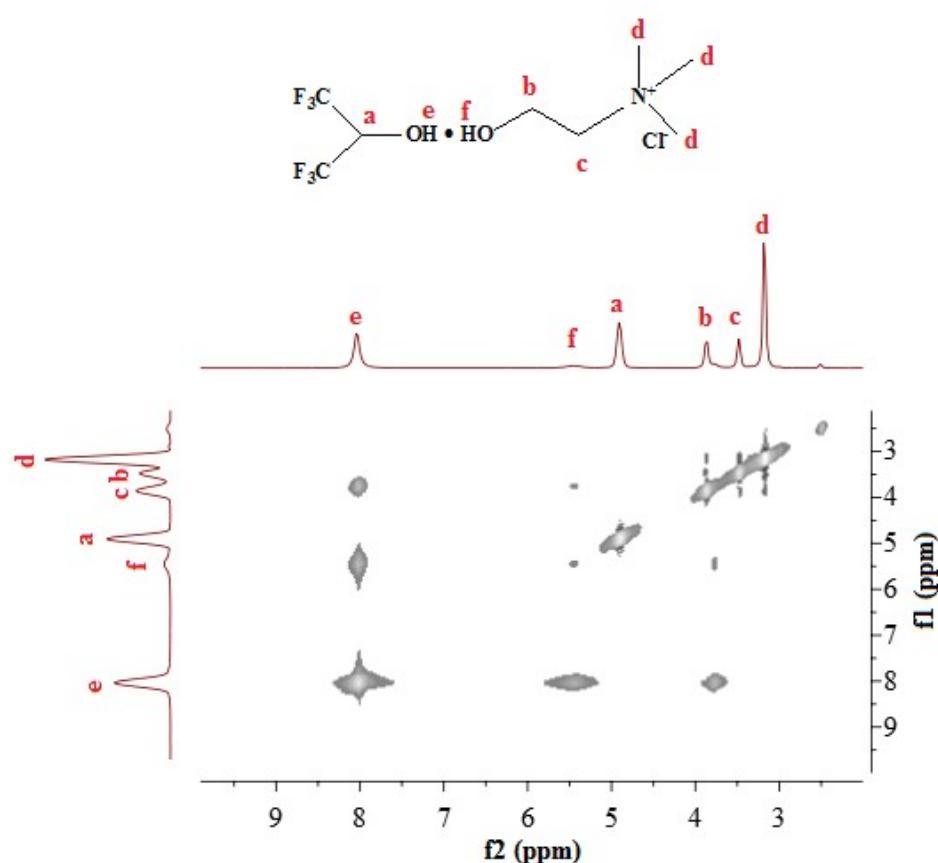


Fig. S1 NOESY spectra of HFIP/ChCl DES.

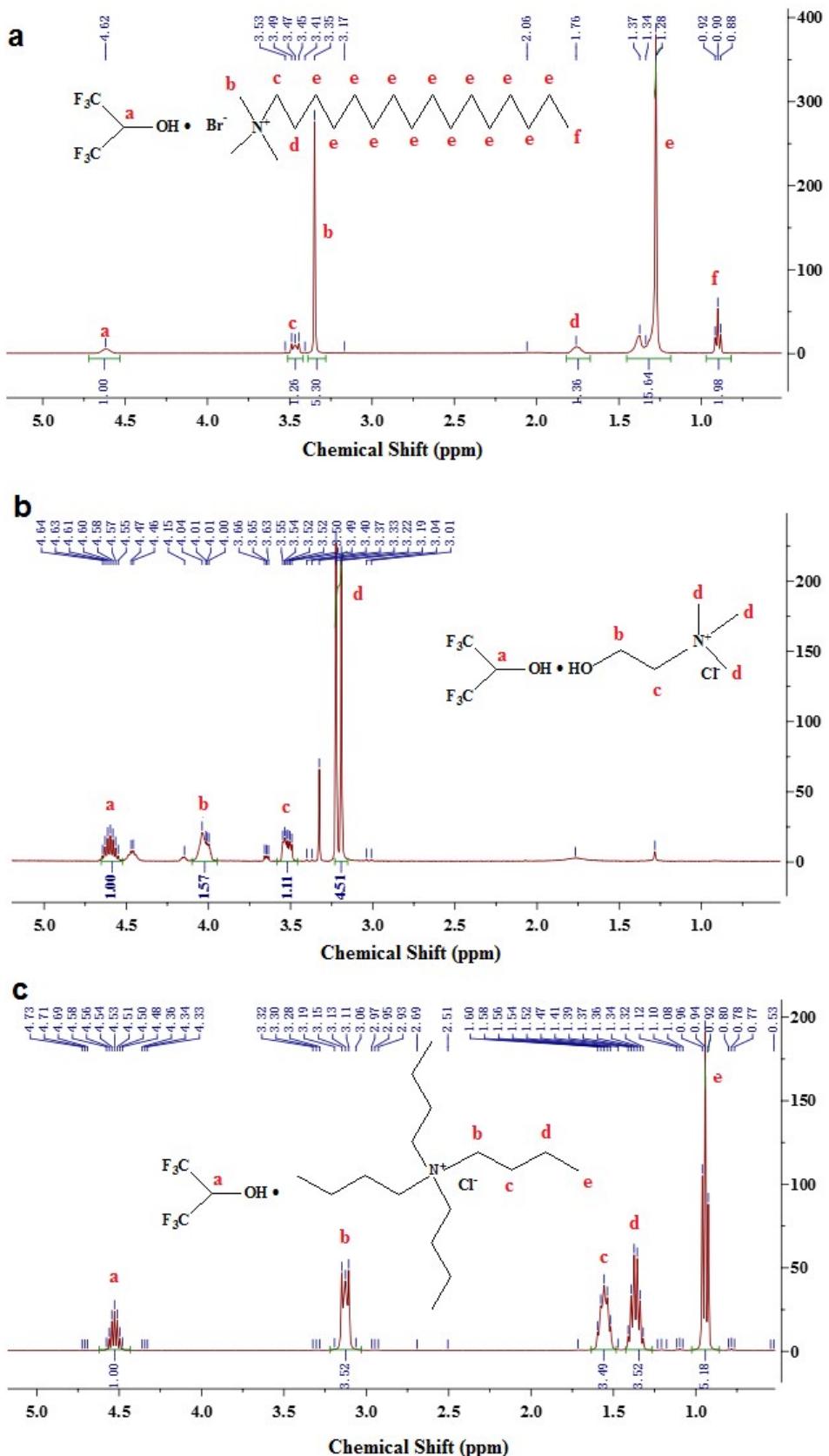


Fig. S2 ^1H -NMR spectra of: (a) HFIP-CTAB, (b) HFIP-ChCl and (c) HFIP-TBAC.

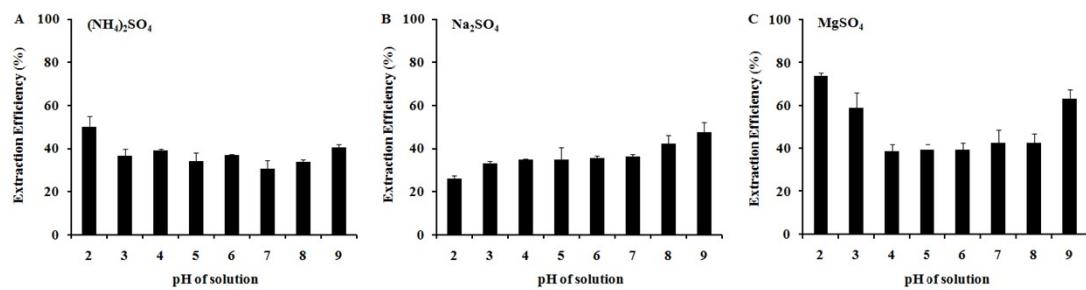


Fig. S3 The effects of sample pH on the extraction of BSA in HFIP-ChCl- $(\text{NH}_4)_2\text{SO}_4$, HFIP-ChCl- Na_2SO_4 and HFIP-ChCl- MgSO_4 systems.

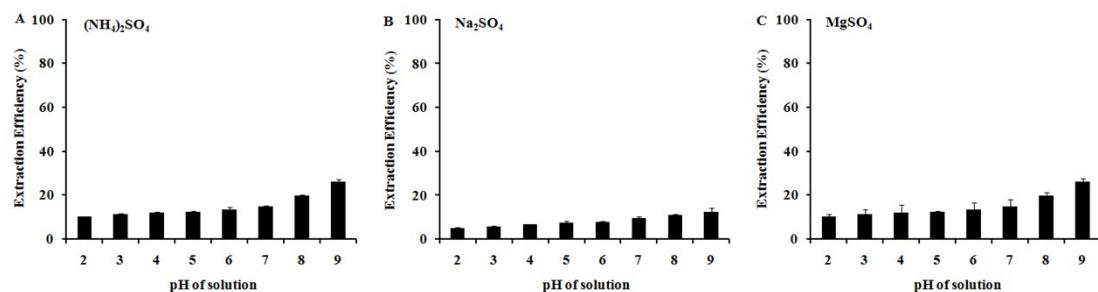


Fig. S4 The effects of sample pH on the extraction of RNA in HFIP/ChCl- $(\text{NH}_4)_2\text{SO}_4$, HFIP/ChCl- Na_2SO_4 and HFIP/ChCl- MgSO_4 systems.

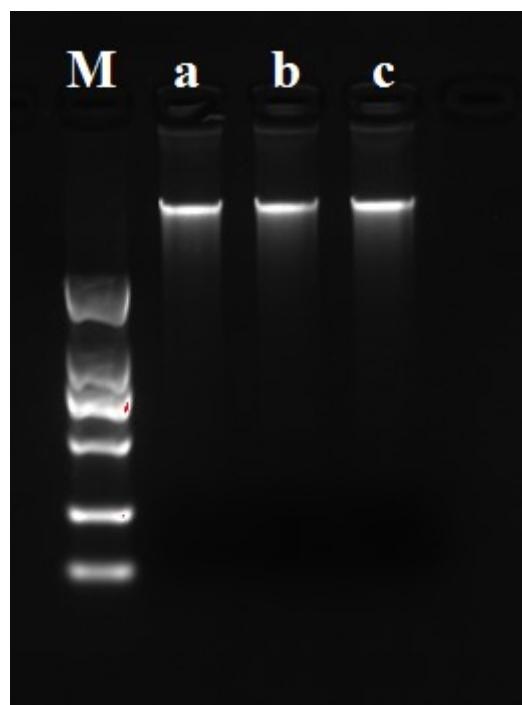


Fig. S5 Agarose gel electrophoresis of the DNA recovered from back extraction using HFIP/ChCl-(NH₄)₂SO₄ system (a), HFIP/ChCl-Na₂SO₄ system (b) and HFIP/ChCl-MgSO₄ system (c). Lane M represents the DNA molecular weight marker 2K.

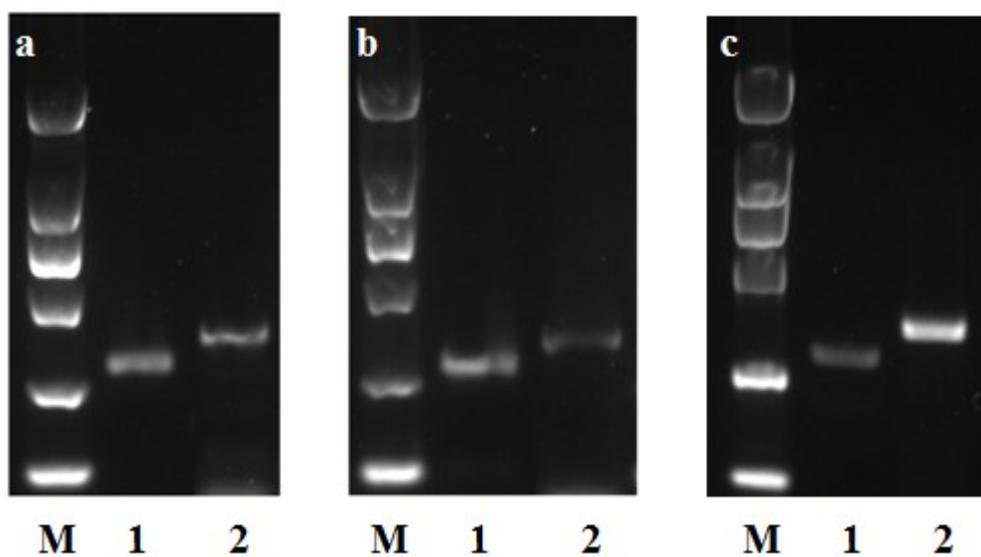


Fig. S6 PCR amplification of *TP53* (lane 1) and *EGFR* (lane 2) sequences from human whole blood using HFIP/ChCl-(NH₄)₂SO₄ system (a), HFIP/ChCl-Na₂SO₄ system (b) and HFIP/ChCl-MgSO₄ system (c). Lane M represents the DNA molecular weight marker 2K.

Table S1 The DNA partitioning behavior in HFIP-based DES systems

| HBA | Inorganic salts | Two-phase formation | Two-phase formation after back extraction | Extraction efficiency of DNA after back extraction |
|------|---|---------------------|---|--|
| ChCl | (NH ₄) ₂ SO ₄ | √ | √ | a |
| | K ₂ HPO ₄ | √ | √ | b |
| | KH ₂ PO ₄ | √ | ✗ | - |
| | Na ₂ CO ₃ | √ | √ | b |
| | Na ₂ HPO ₄ | √ | √ | b |
| | Na ₂ SO ₄ | √ | √ | a |
| | MgSO ₄ | √ | √ | a |
| TBAC | (NH ₄) ₂ SO ₄ | √ | √ | b |
| | K ₂ HPO ₄ | √ | √ | b |
| | KH ₂ PO ₄ | √ | √ | b |
| | Na ₂ CO ₃ | √ | √ | b |
| | Na ₂ HPO ₄ | √ | √ | b |
| | Na ₂ SO ₄ | √ | √ | b |
| | MgSO ₄ | √ | √ | b |
| CTAB | (NH ₄) ₂ SO ₄ | √ | ✗ | - |
| | K ₂ HPO ₄ | ✗ | - | - |
| | KH ₂ PO ₄ | √ | ✗ | - |
| | Na ₂ CO ₃ | ✗ | - | - |
| | Na ₂ HPO ₄ | ✗ | - | - |
| | Na ₂ SO ₄ | √ | ✗ | - |
| | MgSO ₄ | √ | √ | b |

√ The systems can separate into two phases.

✗ The systems cannot be separated into two phases.

a The extraction efficiency of DNA was more than 50%.

b The extraction efficiency of DNA was less than 20%.

Table S2 The primers of genes in this study.

| | | | |
|-------------|--------|----------------|----------------------|
| <i>P53</i> | 274 bp | Forward primer | GTCCCAAGCAATGGATGATT |
| | | Reverse primer | ACTGACCGTGCAAGTCACAG |
| <i>EGFR</i> | 335 bp | Forward primer | AGACGGGAAATTCACACCAG |
| | | Reverse primer | CTGTAAGAGGCAGGGCTTG |