Supporting information:

An optimized method for NMR-based plant seed metabolomic analysis with maximized polar metabolite extraction efficiency, signal-to-noise ratio, and chemical shift consistency

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Figure S1. Extraction procedure for mungbean seed metabolites in NMR-based metabolomic analysis

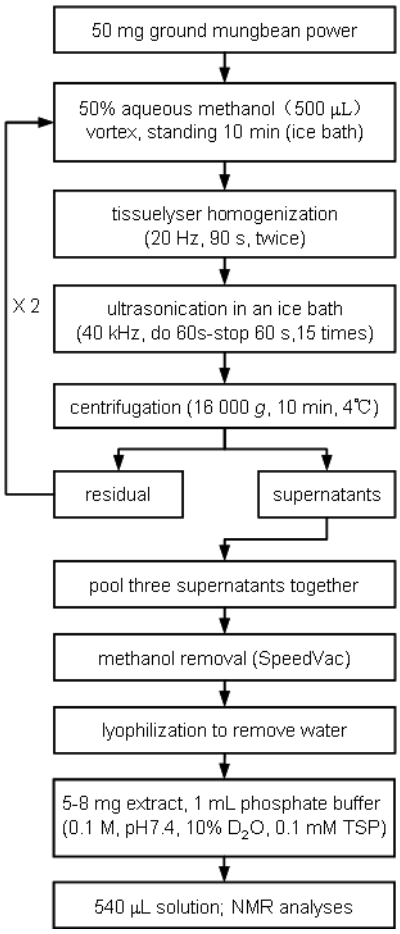

Figure S2. Extraction rates (ER) from aqueous methanol (A) and aqueous acetonitril (B) extraction for mungbean seeds (with no significant inter-group difference).

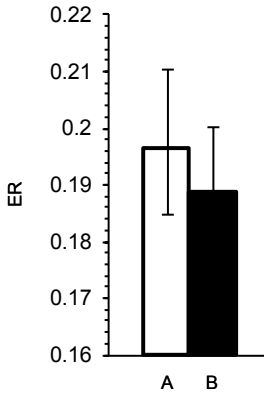

Figure S3 ^{1}H NMR spectra for mungbean extracts from four cell-breaking methods, namely, no treatment (A), tissuelyser homogenization (B), ultrasonication (C) and the combined tissuelyser homogenization with ultrasonication (D).

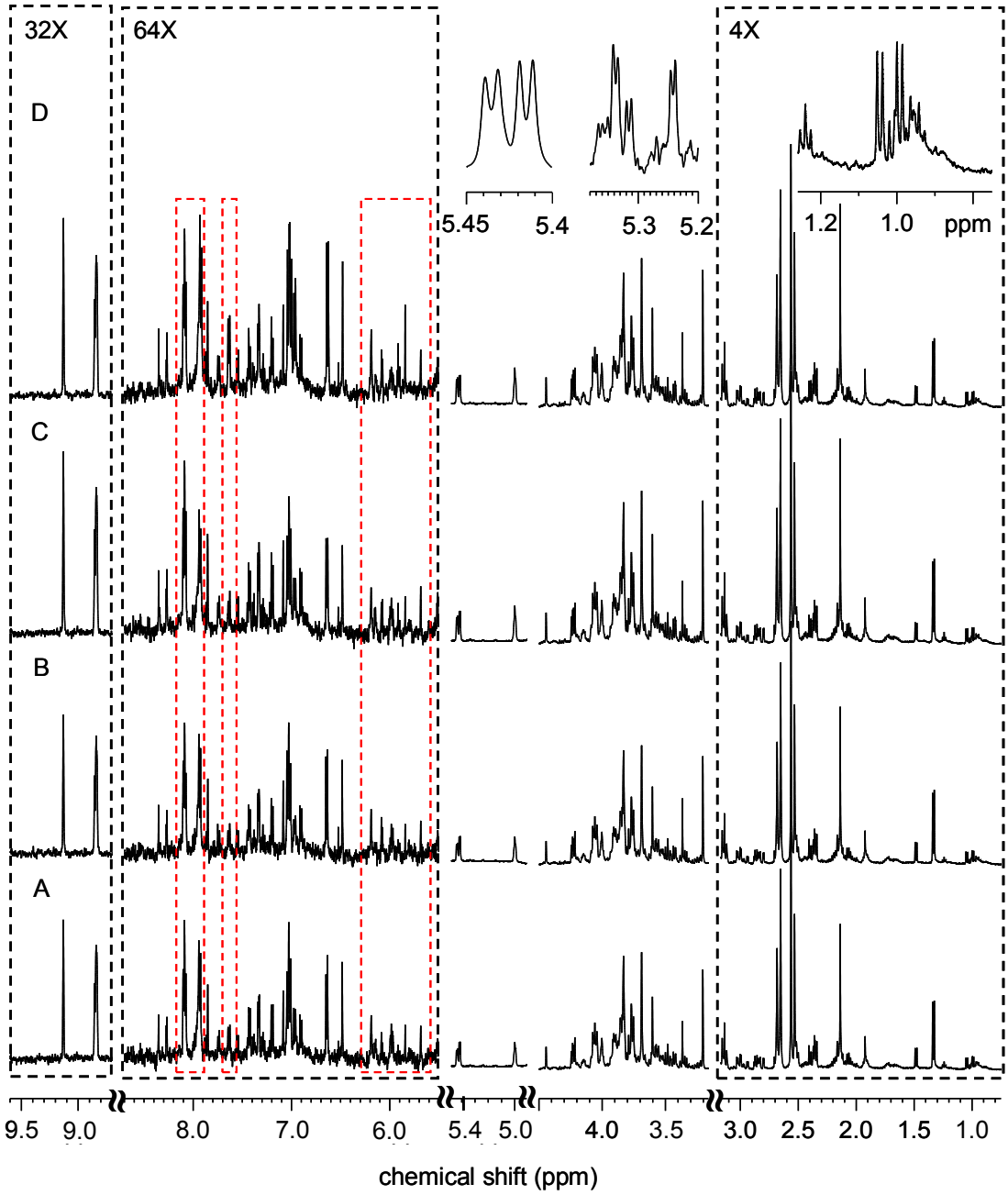

Figure S4 Comparative OPLS-DA results for mungbean extracts from three cell-breaking methods, namely, tissuelyser homogenization (B), ultrasonication (C) and the combined tissuelyser homogenization with ultrasonication (D). (a) R^2X=0.46, Q^2=0.52, p=0.03; (b) R^2X=0.47, Q2=0.49, p=0.02; (c) R^2X=0.38, Q^2=-0.23, p=1

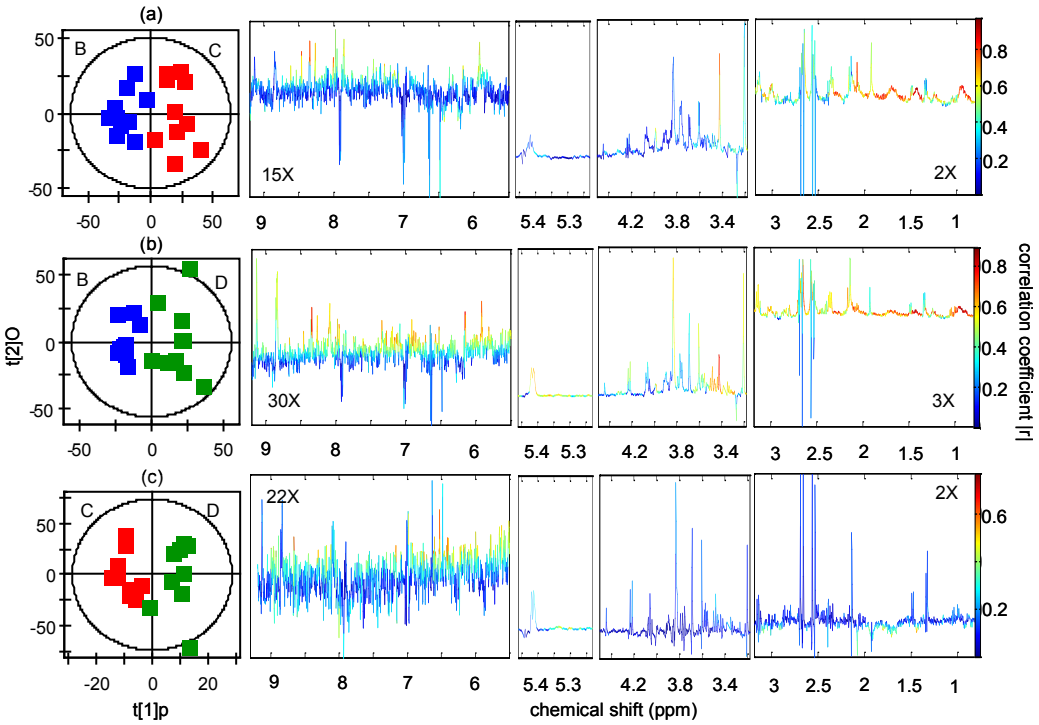

Figure S5. Extraction rates (ER) from six different tissue-to-solvent ratios and four times extractions. Groups with different letters had significant difference ($p<0.05$). E1, E2, E3 and E4 were extracts from the first, second, third and fourth extraction, respectively; E1234 was the pooled extracts from the first four extractions. A, B, C, D, E and F represented the tissue-to-solvent ratios (mg:μL) of 1:5, 1:7.5, 1:10, 1:15, 1:20 and 1:30, respectively.

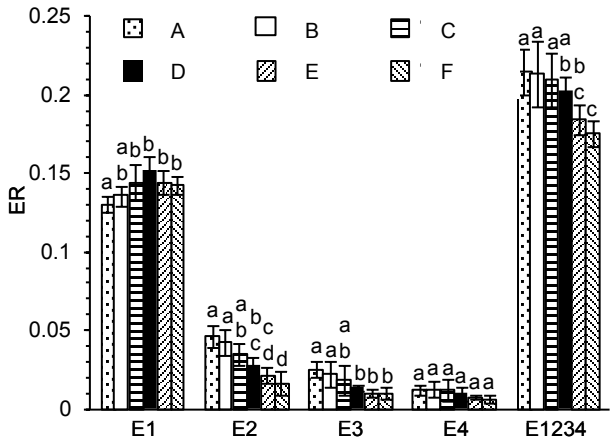

Figure S6. Signal-to-noise ratios (SNR) for citrate in mungbean seed extracts as a function of the extract-to-buffer ratios in NMR analysis.

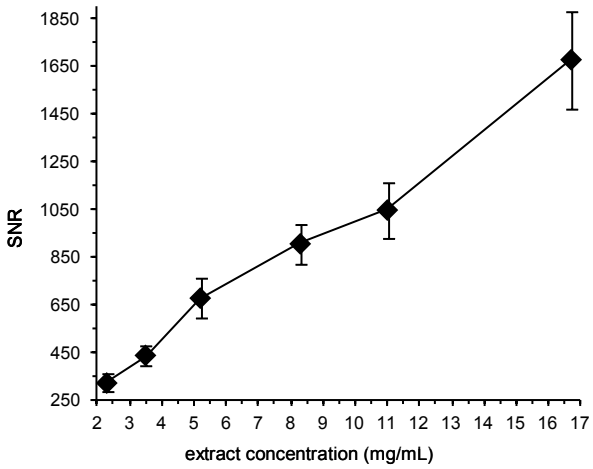

Figure S7. OPLS-DA results for extracts of mungbean seeds germinated for 16 (A) and 18 hrs (B) which resulted from partially relaxed spectra (a: $R^2X=0.61$, $Q^2=0.99$, $p=1.3e^{-15}$) and from fully relaxed spectra(b: $R^2X=0.83$, $Q^2=0.99$, $p=2.8e^{-14}$), respectively.