SUPPLEMENTARY MATERIAL

Perfluorocarbon nanoemulsions for simultaneous delivery of oxygen and antioxidants during machine perfusion supported organ preservation

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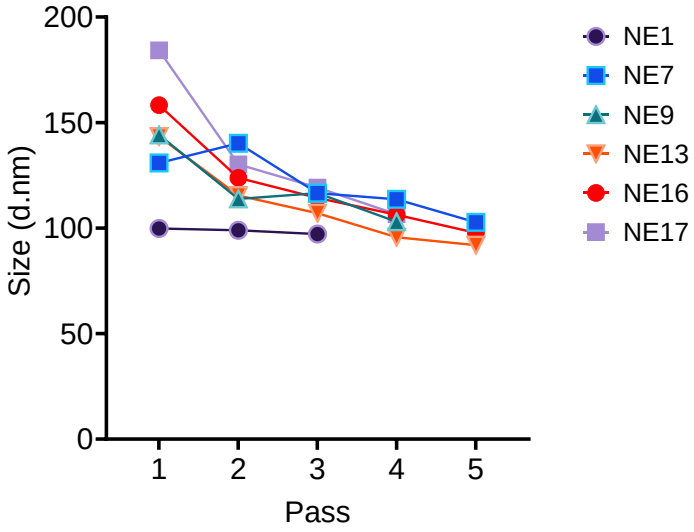


Figure S1. Particle size of discrete passes for a representative NE batch from each production scale.

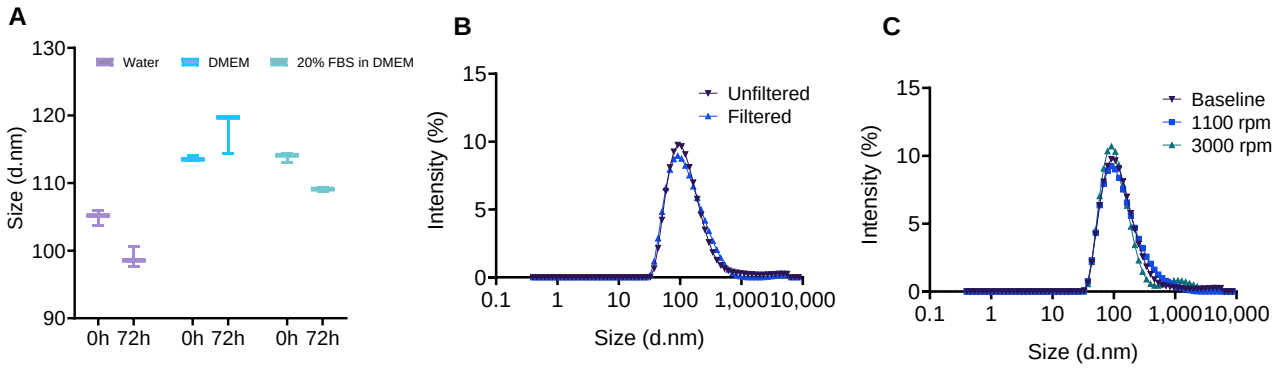


Figure S2. Colloidal stability of DF-NE (1000 mL): (A) after 72h incubation in serum and serum-free medium; (B) after filtration through 0.44 µm; (C) after centrifugation at 1100 and 300 rpm.

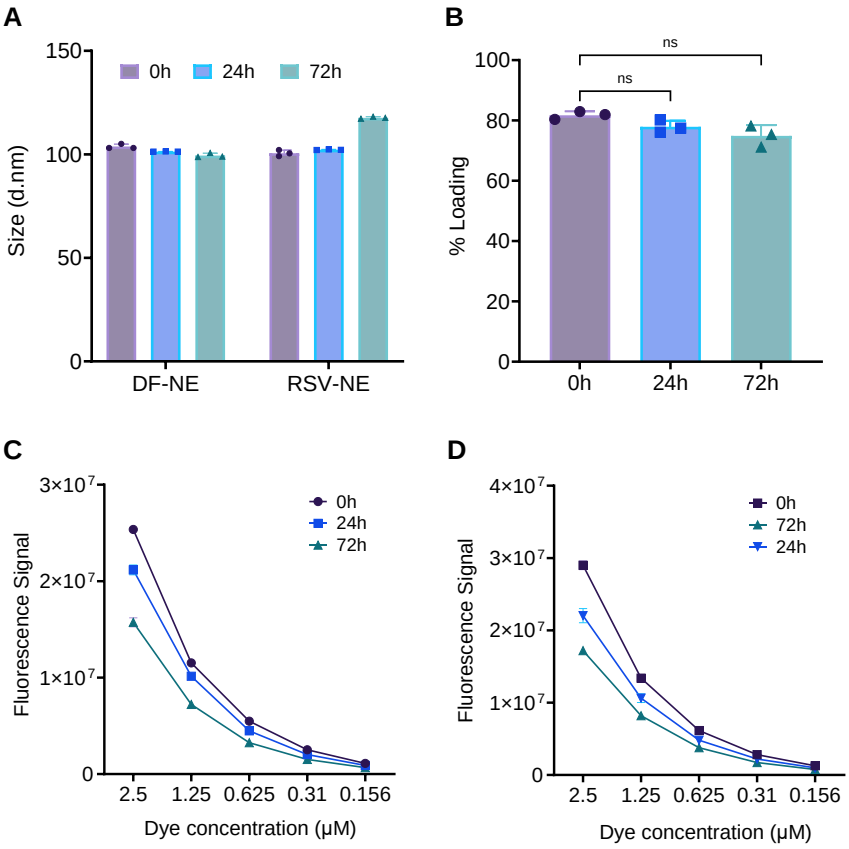


Figure S3. Accelerated stability studies of DF-NE and RSV-NE at 37°C for 24 and 72 hours: (A) particle size; (B) % drug loading; fluorescence signal of (C) DF-NE and (D) RSV-NE. Each datapoint represents $n = 3$. One-way ANOVA with Dunnett's multiple comparison, unpaired t-test, ns not significant.

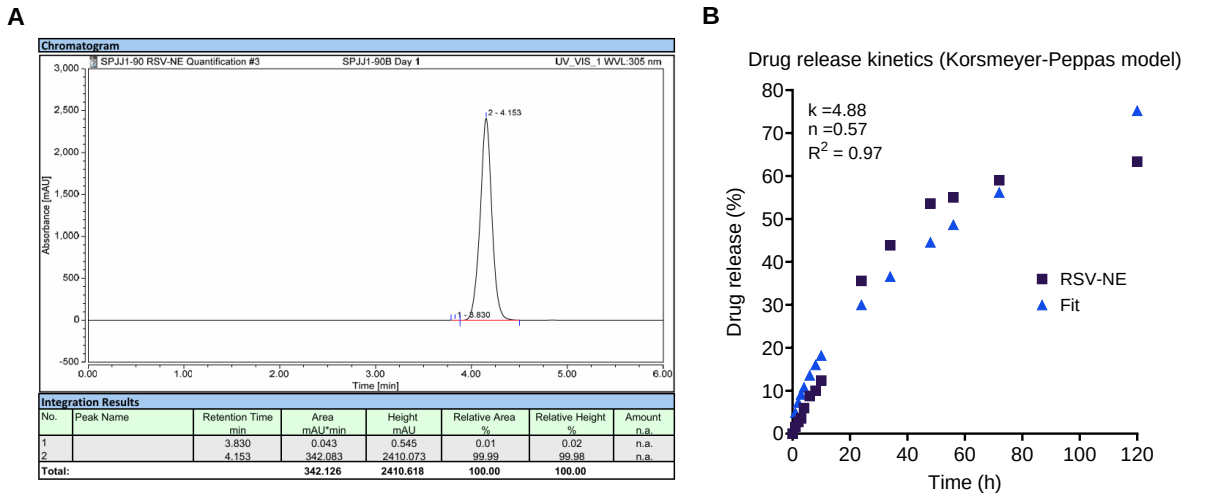


Figure S4. HPLC chromatogram and drug release kinetics model. (A) Representative image of HPLC chromatogram of resveratrol quantified from the resveratrol-loaded nanoemulsion sample; (B) drug release kinetics (Korsmeyer-Peppas model) with an R2 value of 0.97 and n=0.57, following non-Fickian diffusion.

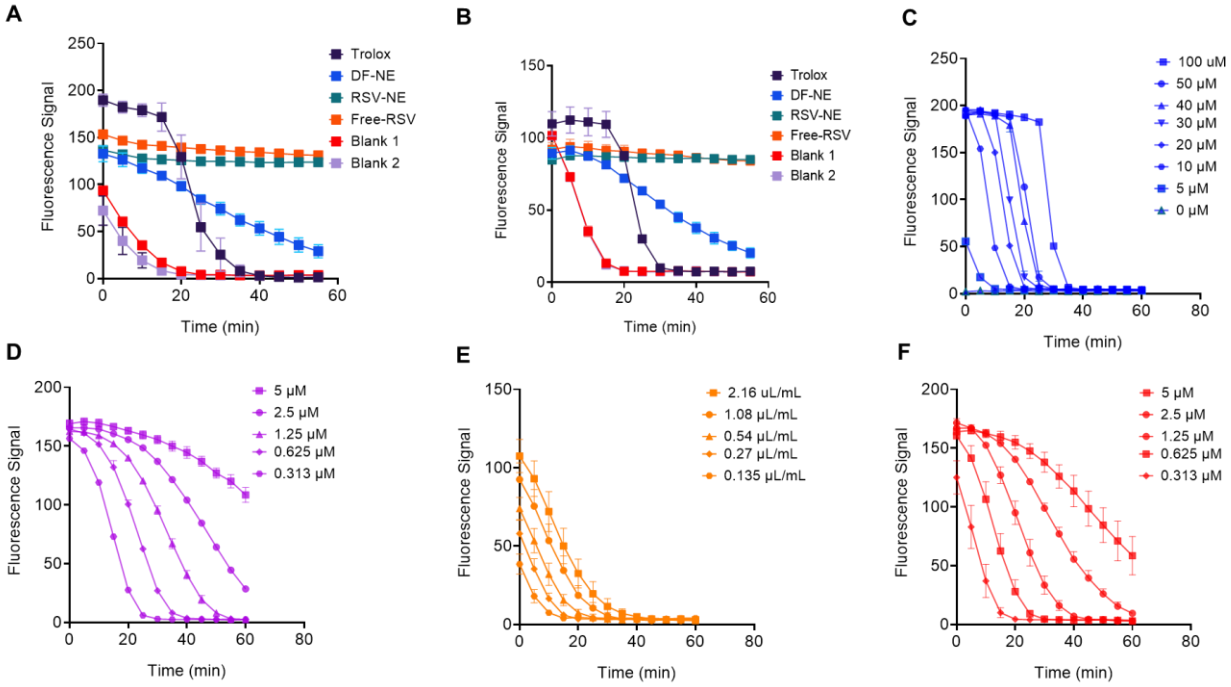


Figure S5. In-vitro Oxiselect™ Oxygen Radical Antioxidant Capacity (ORAC) activity assay. (A, B) AAPH radical-mediated fluorescence decay curves of Trolox, DF-NE, RSV-NE, Free-RSV, Blank 1 (Assay diluent), and Blank 2 (Water) in two independent assays. Inhibition of AAPH radical-mediated fluorescence decay curve by Trolox (C), Free-RSV (D), DF-NE (E), and RSV-NE (F) at different concentrations. Data represented as mean ± SD (n = 3).

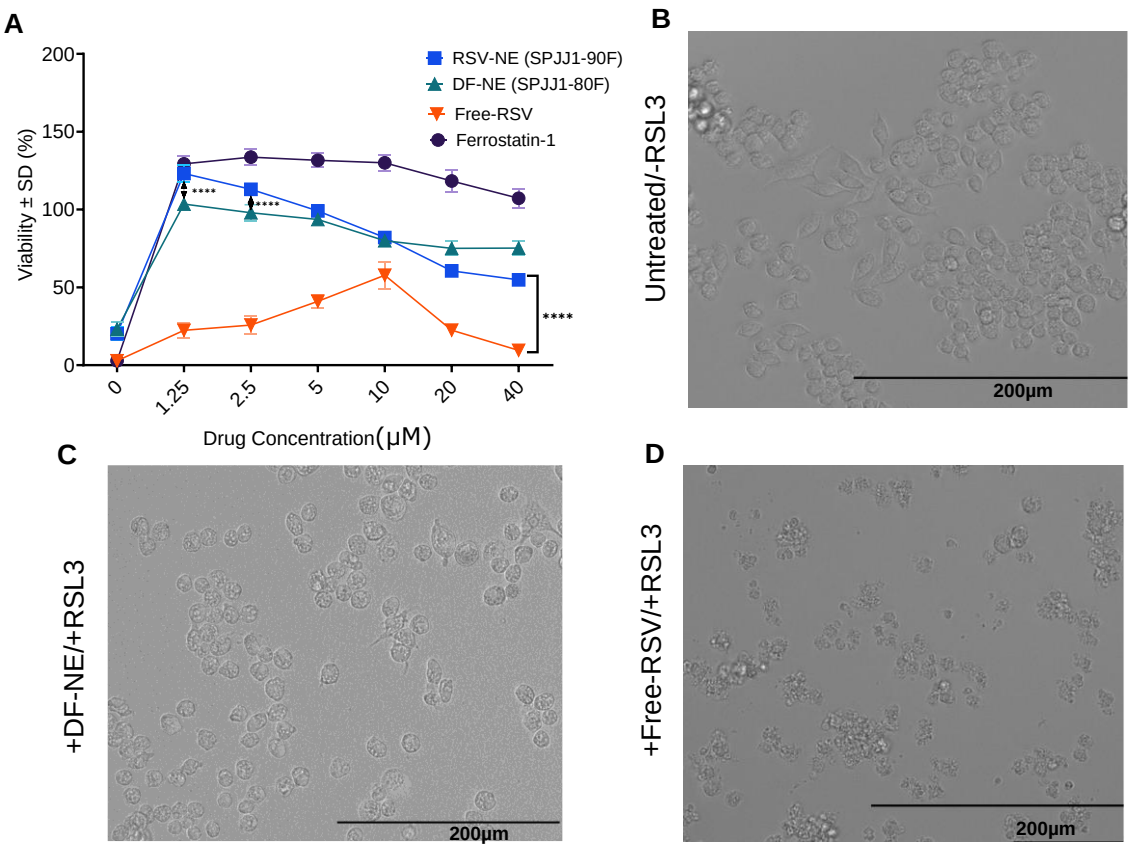


Figure S6. Replicated ferroptosis inhibition assay and EVOS microscopic analysis. (A) Dose-dependent ferroptosis inhibition in RSL3-induced RAW 264.7 macrophages by RSV-NE, volume-matched DF-NE, drug concentration-matched Free-RSV, and Ferrostatin-1. The % macrophage viability was assessed post-exposure to RSL3 (ferroptosis inducer) for 6 h. EVOS microscopic images of non-activated cells (B) and RSL3-activated macrophage cells exposed to complete cell culture medium (DF-NE) (C) and Free-RSV (D) at 20x magnification. Data represent mean ± SD (n=6). Two-way ANOVA with Tukey's multiple comparison, unpaired t-test, **** p<0.0001.