Supplementary Information

Solvent-Mediated Organocatalytic Browning of Biogenic Indoles Enables the Formation of Zwitterionic Nanoparticles

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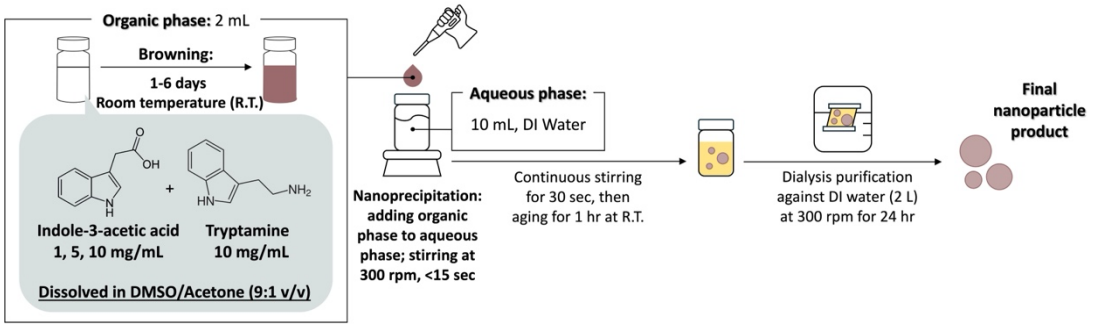

Scheme S1. Schematic overview illustrating the transformation of biogenic indoles into browned nanoparticles through solvent-mediated organocatalytic browning. The workflow outlines three major experimental stages: (i) modulation of browning extent in DMSO/acetone (9:1, v/v) by varying indole composition and reaction time; (ii) formation of colloidal dispersions via nanoprecipitation into water; and (iii) purification, optimization, and physicochemical characterization of the resulting nanoparticles.

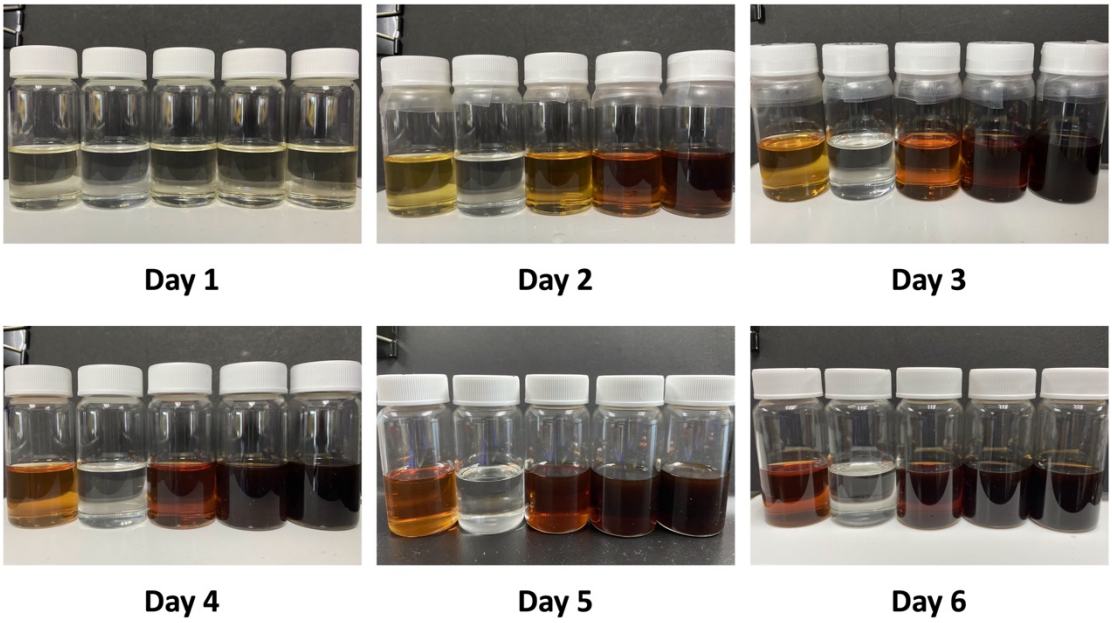

Fig. S1. Photographic record of the browning process in the organic phase over time. Representative images showing the color evolution of reaction mixtures in DMSO/acetone (9:1, v/v) at room temperature. Photographs were taken daily for five experimental groups (left to right): tryptamine alone (10 mg mL^{-1}), I3AA alone (10 mg mL^{-1}), and tryptamine (10 mg mL^{-1}) combined with 1, 5, and 10 mg mL^{-1} of I3AA, respectively. The progressive darkening reflects the extent of solvent-mediated browning as a function of I3AA concentration and reaction time.

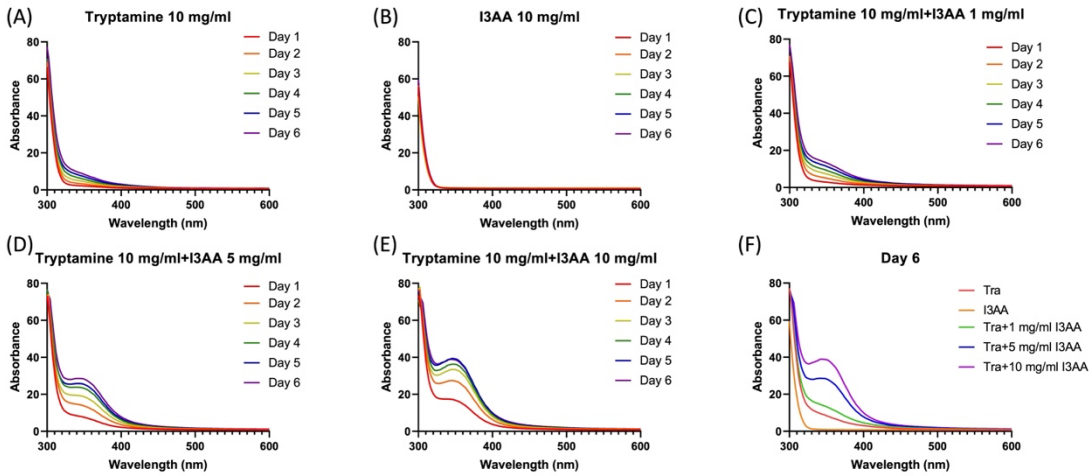

Fig. S2. UV–visible spectral analysis of the browning reactions shown in Figure S1. (A–E) Time-evolved UV–visible absorption spectra for each reaction group, corresponding to tryptamine alone, I3AA alone, and tryptamine combined with 1, 5, and 10 mg mL^{-1} of I3AA, respectively. Each panel displays six spectra recorded from day 1 to day 6. (F) Comparative overlay of the spectra collected on day 6 across all five reaction groups.

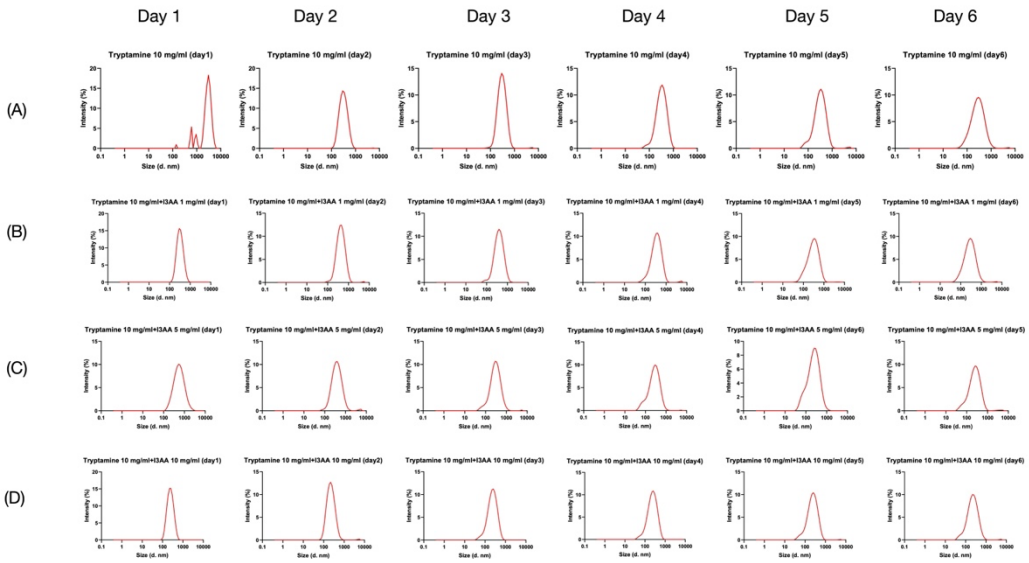

Fig. S3. Hydrodynamic size distribution profiles of nanoprecipitated indole-derived particles.

Dynamic light scattering (DLS) measurements showing particle size distributions of dispersions obtained from organic phases reacted for 1–6 days. (A) Tryptamine alone (10 mg mL^{-1}); (B–D) tryptamine (10 mg mL^{-1}) combined with 1, 5, and 10 mg mL^{-1} of I3AA, respectively.

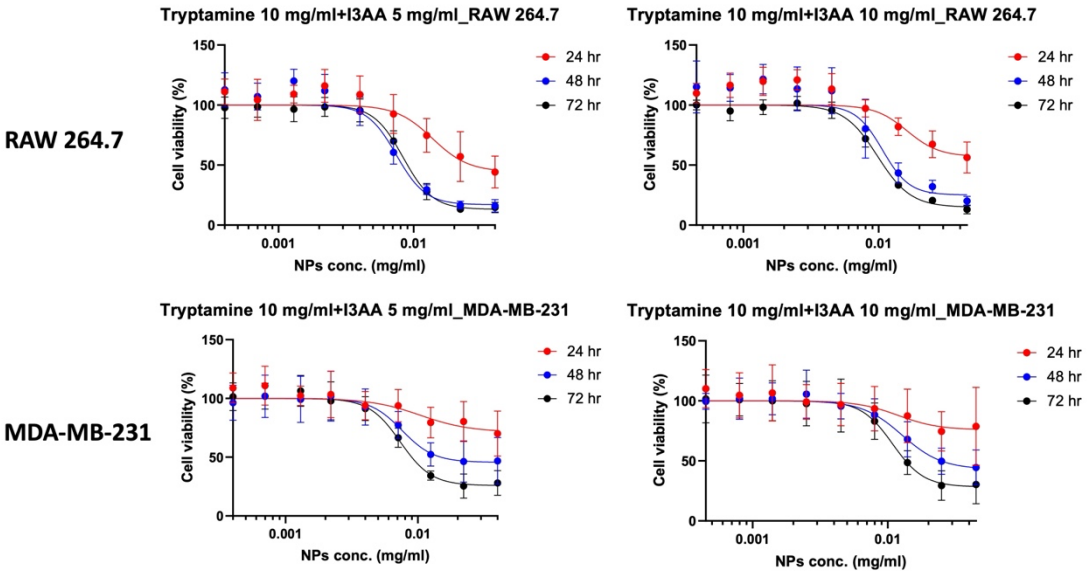

Fig. S4. Cytotoxicity profiles of browned indole-derived nanoparticles.
Cell viability (mean ± SD, n = 3) of RAW 264.7 cells (upper panel) and MDA-MB-231 cells (lower panel) after 24, 48, and 72 h incubation with nanoparticle dispersions at varying concentrations. The left panels correspond to formulation F1 (nanoparticles prepared from 10 mg mL^{-1} tryptamine and 5 mg mL^{-1} indole-3-acetic acid, I3AA), and the right panels correspond to formulation F2 (nanoparticles prepared from 10 mg mL^{-1} tryptamine and 10 mg mL^{-1} I3AA).

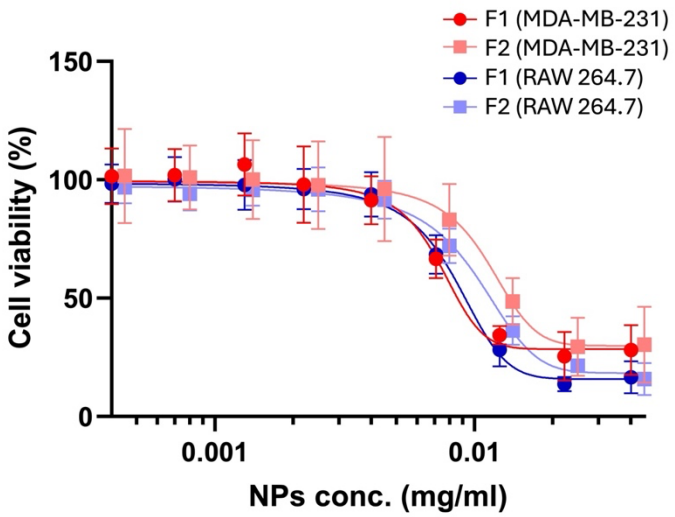

Fig. S5. Cytotoxicity comparison of browned indole-derived nanoparticles.
Cell viability (mean ± SD, n = 3) after 72 h incubation with nanoparticle dispersions at varying concentrations. Formulation F1: nanoparticles prepared from 10 mg mL^{-1} tryptamine + 5 mg mL^{-1} indole-3-acetic acid (I3AA). Formulation F2: nanoparticles prepared from 10 mg mL^{-1} tryptamine + 10 mg mL^{-1} I3AA.

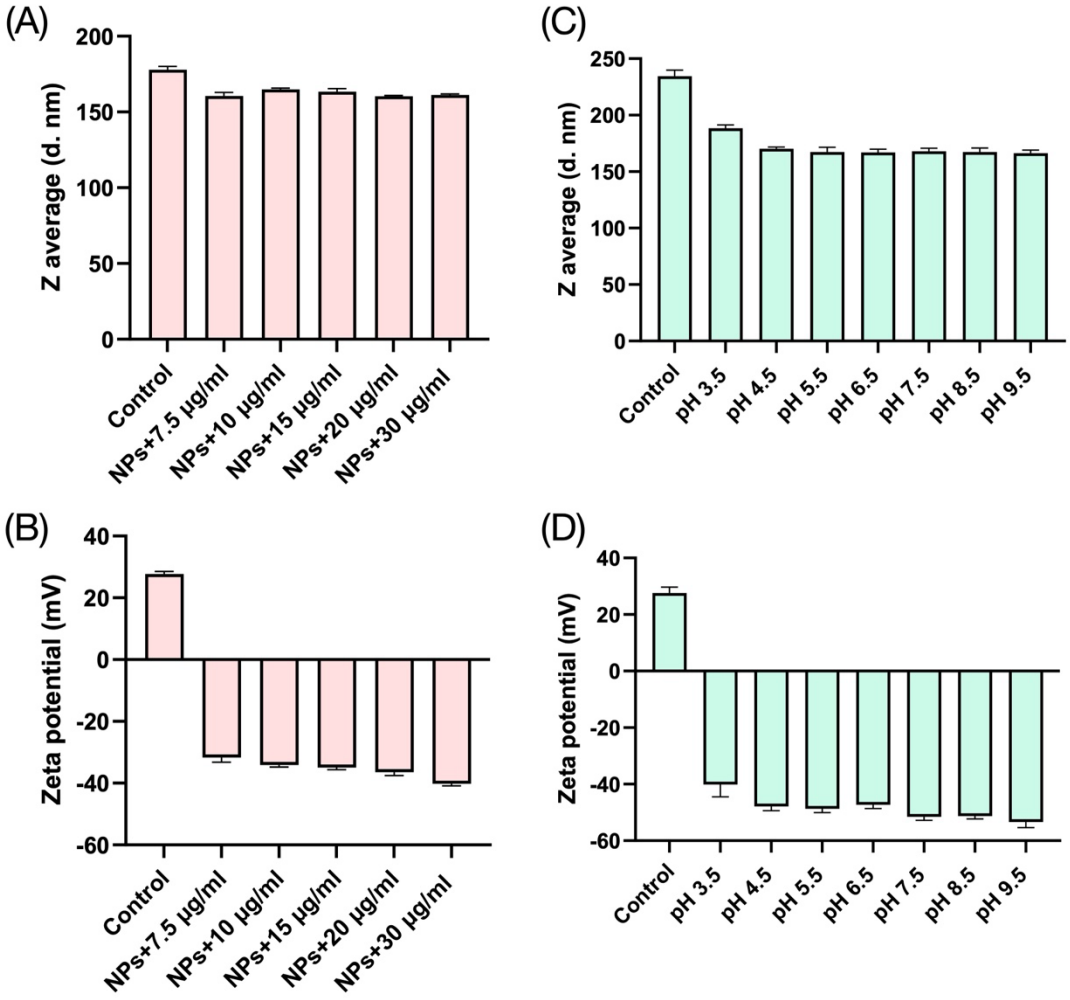

Fig. S6. Characterization of nanoparticle–polynucleotide electrostatic complexation. Changes in hydrodynamic diameter (upper panels) and zeta potential (lower panels) of browned indole-derived nanoparticles upon association with the model oligonucleotide G3139. (A, B) Effects of increasing G3139 concentration on nanoparticle size and surface charge. (C, D) Effects of solution pH on the physicochemical properties of nanoparticle–G3139 complexes.

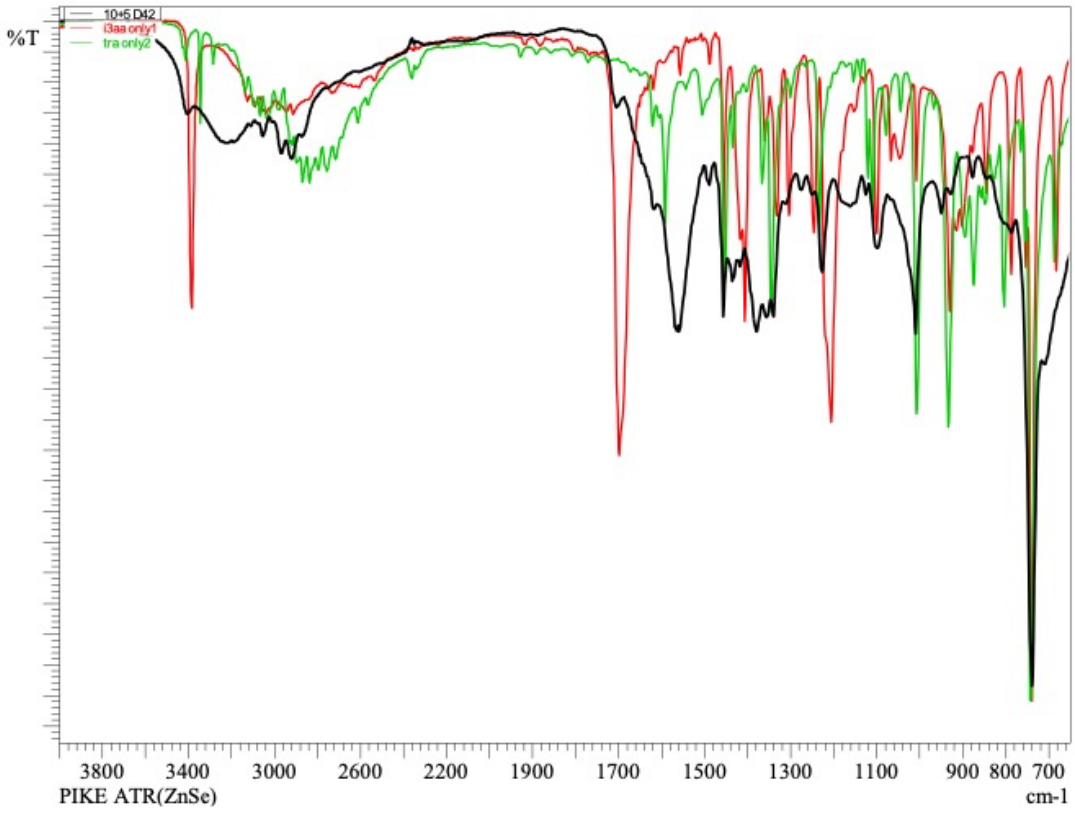

Fig. S7. ATR-FTIR spectra of authentic tryptamine (green), I3AA (red), and browned indole-derived nanoparticles (black).

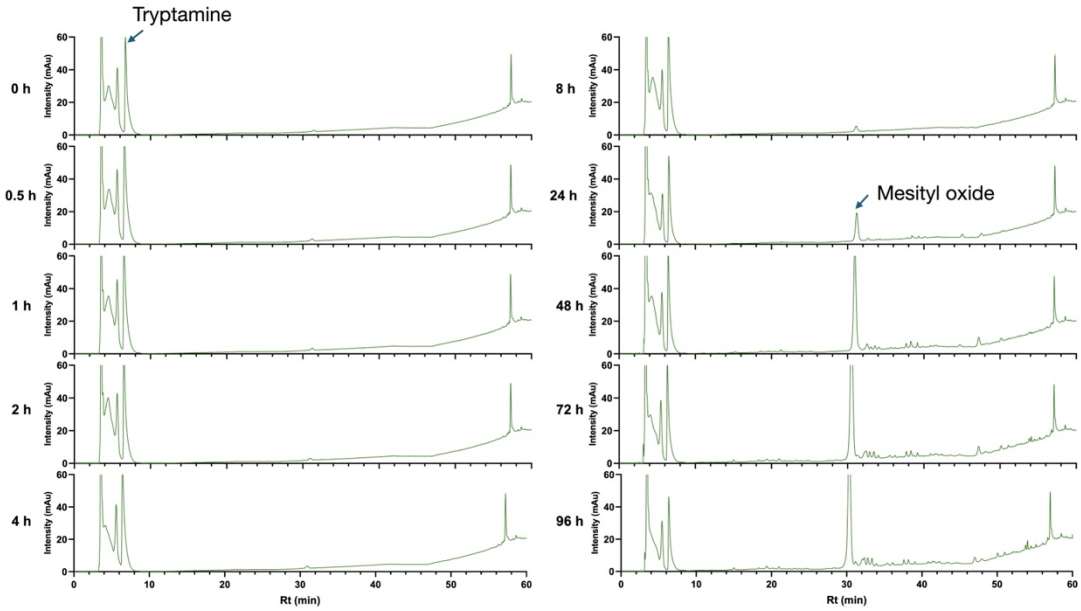

Fig. S8. Time-evolved HPLC chromatograms of the browning reaction in the tryptamine-only system.

Chromatographic profiles of the organic phase containing tryptamine (10 mg mL^{-1}) undergoing solvent-mediated browning in DMSO/acetone (9:1, v/v) over 0–96 h. The emergence of the mesityl oxide (MO) peak at 24 h indicates delayed onset of acetone self-condensation in the absence of indole-3-acetic acid (I3AA), contrasting with the immediate MO formation observed in the mixed-indole system (cf. Figure 8, main text).

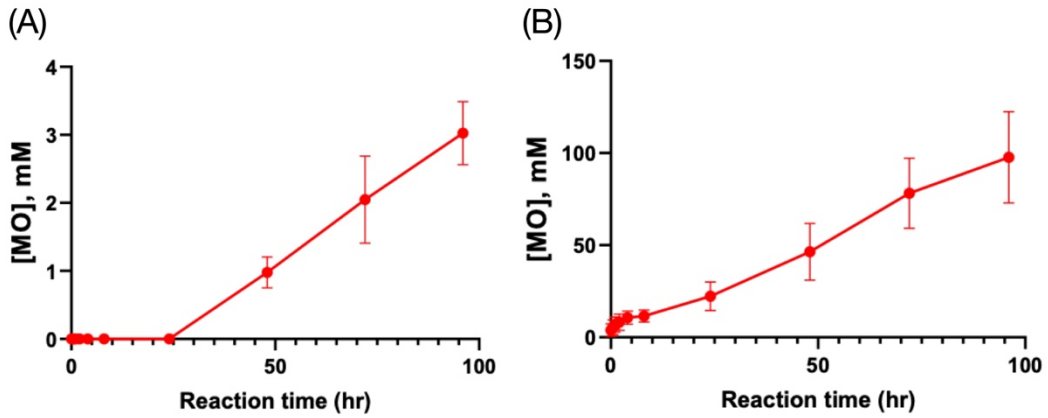

Fig. S9. Quantitative comparison of mesityl oxide (MO) formation over time in the tryptamine-only (A) and the mixed-indole (B) systems.

Table S1. Physicochemical properties of browned indole-derived nanoparticles

Formulation	pH	Hydrodynamic diameter (nm)	Zeta potential (mV)	PDI
F1	6.8	206.3±0.6	46.2±1.1	0.16±0.02
F2	6.8	194.1±2.2	44.3±1.1	0.14±0.01

F1: Tryptamine 10 mg mL^{-1} + I3AA 5 mg mL^{-1}.

F2: Tryptamine 10 mg mL^{-1} + I3AA 10 mg mL^{-1}.