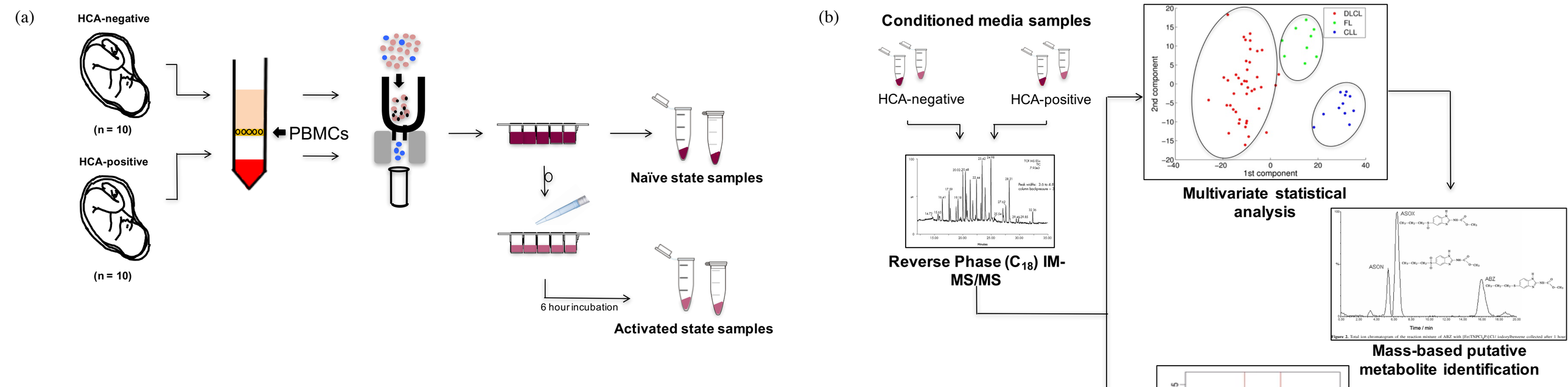


**Figure S1: Experimental design workflow and analysis strategy.**

(a) Peripheral blood was drawn from preterm infants and PBMCs were isolated immediately. Naïve $CD4^+$ T lymphocytes were magnetically isolated on the day of experiment and plated. Conditioned media samples were harvested prior to and 6 hours after SEB/anti-CD28 addition.

(b) Illustration of data acquisition and analysis methods. Conditioned media samples were prepared for exo-metabolomic analysis and analyzed with ultra-performance liquid chromatography-ion mobility-mass spectrometry. Analytical methods were then employed to transition the multidimensional dataset into statistically prioritized metabolic features and putative identifications.

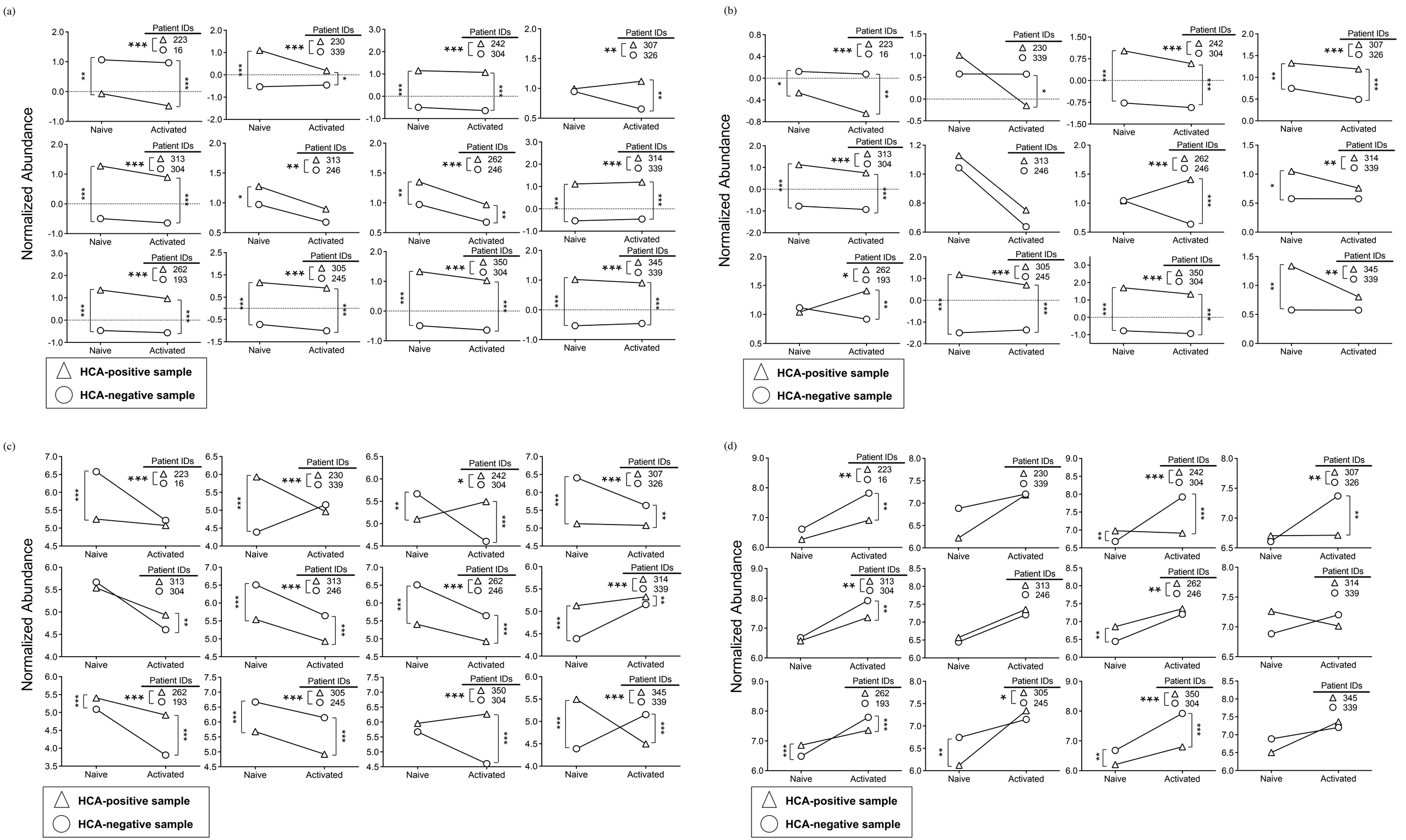


**Figure S2: Pairwise comparisons of identified metabolites' normalized abundance.**

(a) Pairwise comparison of the normalized abundance of 5-HT (*m/z* 177.08, RT 2.57 min) in the naïve and activated states. Each graph depicts the results of 2-way repeated measures ANOVA. HCA-positive samples are represented with triangles while HCA-negative samples are represented by circles. Statistical significance is denoted with stars ($p < 0.05$ = *; $p < 0.01$ = **).

(b) Pairwise comparison of the normalized abundance of 3-HK (*m/z* 225.09, RT 1.25 min) in the naïve and activated states. Each graph depicts the results of 2-way repeated measures ANOVA. HCA-positive samples are represented with triangles while HCA-negative samples are represented by circles. Statistical significance is denoted with stars ($p < 0.05$ = *; $p < 0.01$ = **).

(c) Pairwise comparison of the normalized abundance of DA (*m/z* 176.07, RT 4.63 min) in the naïve and activated states. Each graph depicts the results of 2-way repeated measures ANOVA. HCA-positive samples are represented with triangles while HCA-negative samples are represented by circles. Statistical significance is denoted with stars ($p < 0.05$ = *; $p < 0.01$ = **).

(d) Pairwise comparison of the normalized abundance of LPC (*m/z* 520.35, RT 10.49 min) in the naïve and activated states. Each graph depicts the results of 2-way repeated measures ANOVA. HCA-positive samples are represented with triangles while HCA-negative samples are represented by circles. Statistical significance is denoted with stars ($p < 0.05$ = *; $p < 0.01$ = **).

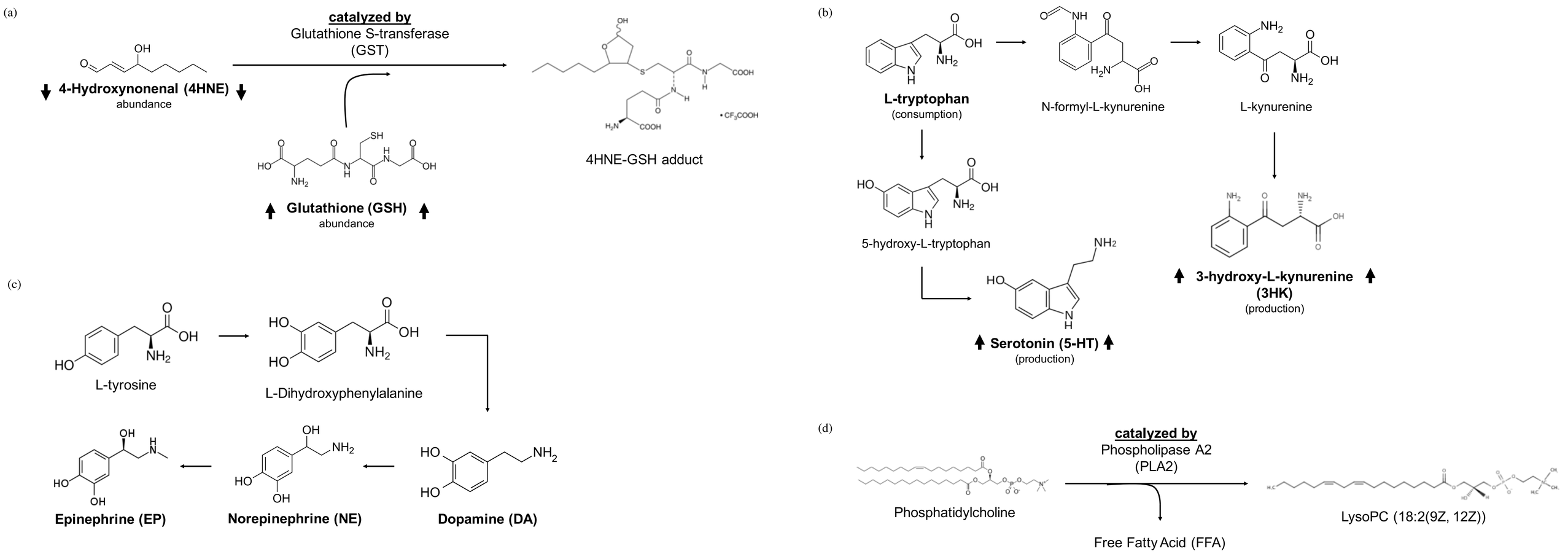


**Figure S3: Pathways depicting biosynthesis and/or degradation of identified metabolites.**

(a) Pathway illustrating 4-HNE detoxification; decreased 4-HNE abundances may result from increased GSH levels.

(b) Pathway illustrating L-tryptophan degradation; elevated levels of 5-HT and 3-HK may indicate up-regulation of L-tryptophan degradation.

(c) Pathway illustrating catecholamine biosynthesis.

(d) Pathway illustrating LysoPC (18:2(9Z, 12Z)) biosynthesis.