Supplemental material

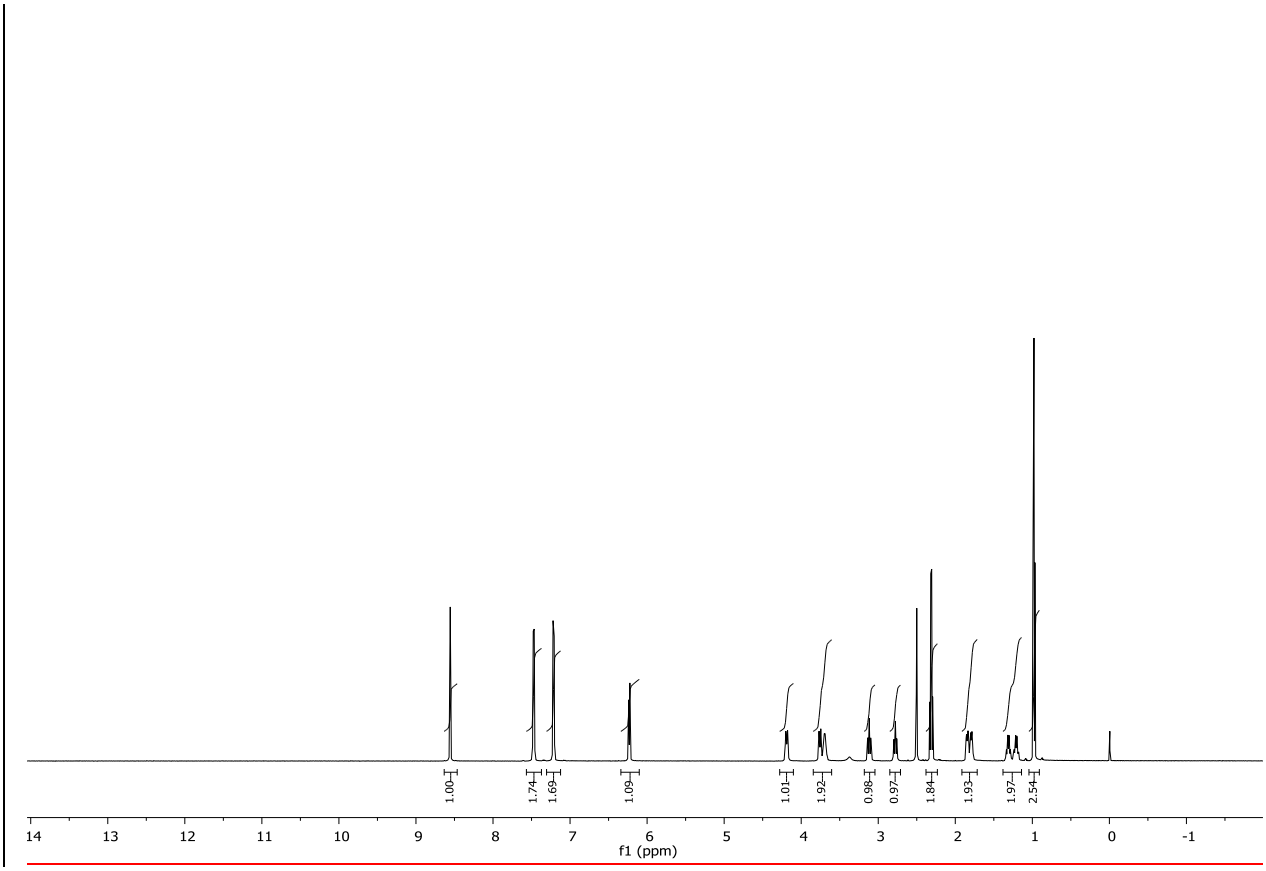

Figure S1. ^{1}H NMR spectrum for TPPU. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.56 (s, 1H), 7.47 (dd, J = 9 and 2 Hz, 2H), 7.21 (d, J = 9 Hz, 2H), 6.23 (d, J = 8 Hz, 1H), 4.19 (d, J = 13 Hz, 1H), 3.76 (d, J = 15 Hz, 1H), 3.72-3.66 (m, 1H), 3.12 (ddd, J = 14, 11, and 3 Hz, 1H), 2.78 (ddd, J = 14, 11, and 3 Hz, 1H), 2.31 (q, J = 7 Hz, 2H), 1.87-1.76 (m, 2H), 1.35-1.17 (m, 2H), 0.98 (t, J = 7 Hz, 3H).

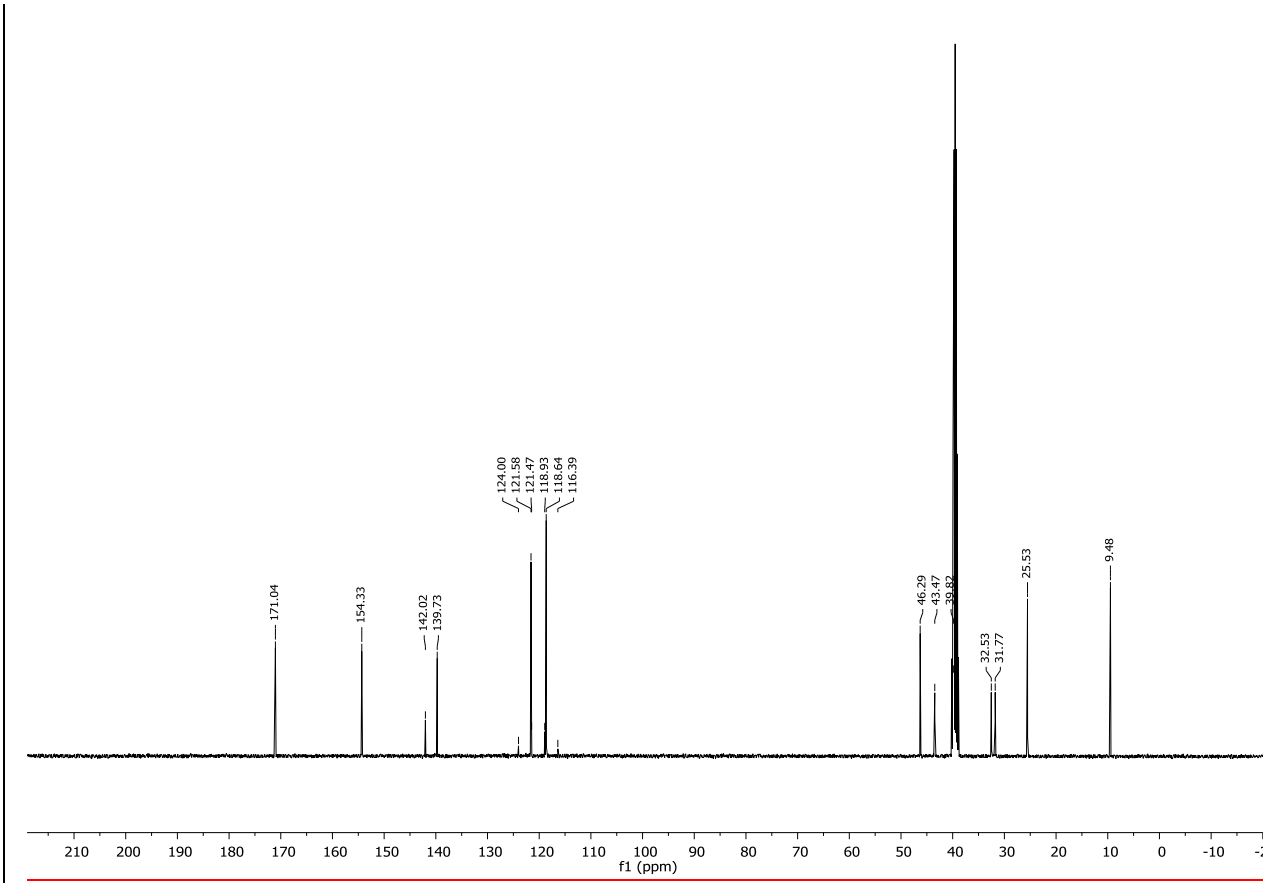

Figure S2. ^{13}C NMR spectrum for TPPU. ^{13}C NMR (100 MHz, DMSO-d_6) δ 171.04, 154.33, 142.02, 139.73, 121.58, 120.20 (q, J = 255 Hz), 118.64, 46.29, 43.47, 39.82, 32.53, 31.77, 25.53, 9.48.

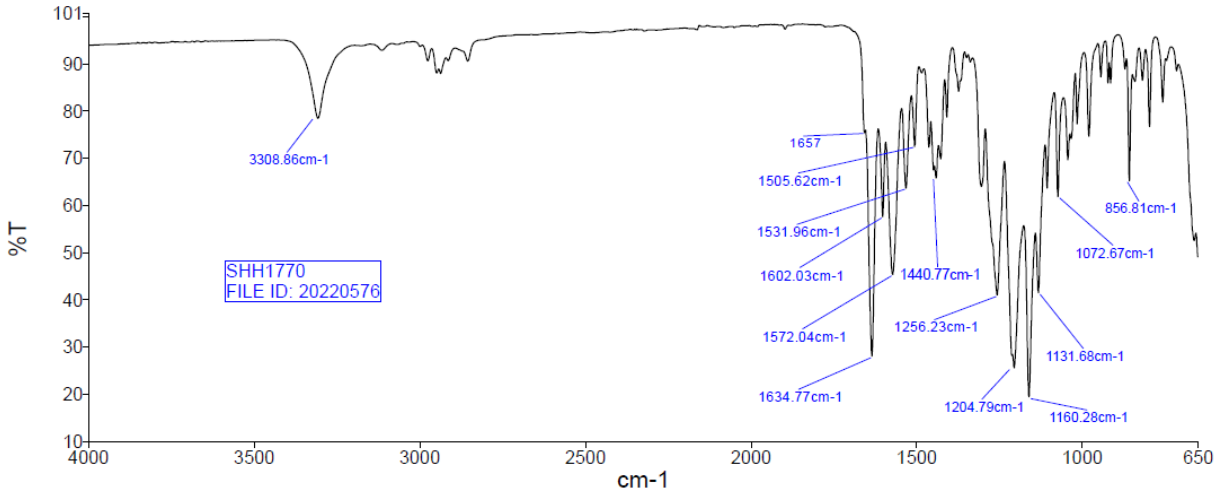

Figure S3. ATR-FTIR spectrum for TPPU. IR (neat) 3308.86, 1634.77, 1602.03, 1572.04, 1531.96, 1505.62, 1440.77, 1303.97, 1256.23, 1204.79, 1160.28, 1131.68, 1105.16, 1072.67, 1042.49, 1014.54, 978.86, 921.03, 856.81, 795.81, 755.92 cm^{-1}.

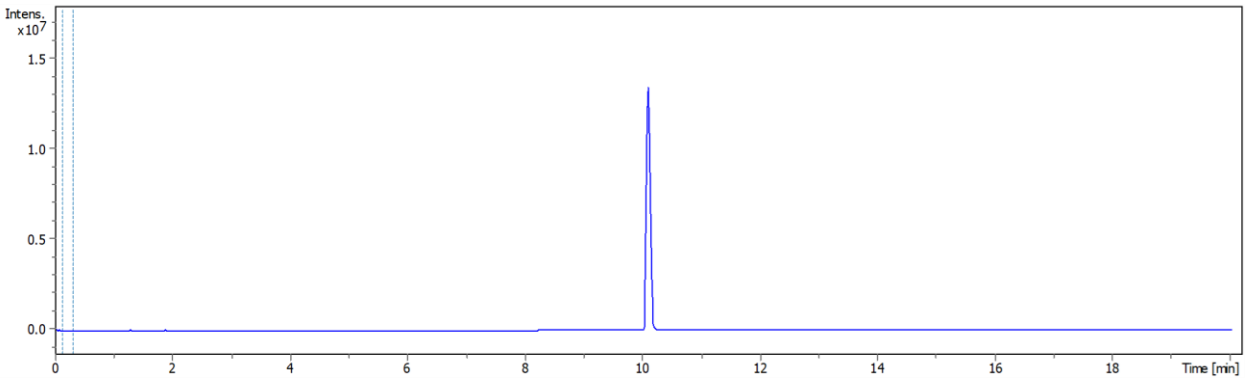

Figure S4. Purity of TPPU based on the total ion chromatogram. Anal. Calcd $C_{16}H_{20}F_3N_3O_3$: C, 53.48; H, 5.61; N, 11.69. Found: C, 53.49; H, 5.38; N, 11.70. Purity was greater than 99%.

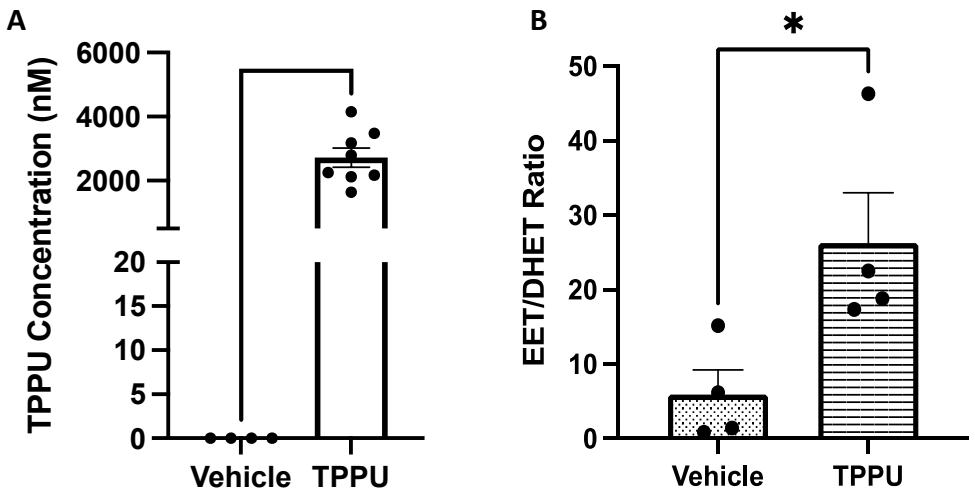

Figure S5. LC/MS analysis of murine blood and plasma. **A.** Peripheral TPPU concentration was assessed using whole blood collected from the tail vein of mice upon the completion of chronic TPPU treatment. Although there was some variability in the detected concentration, at least 1 μM of TPPU was detected in all mice tested (n=4 or 8, $p<0.001$), which is orders of magnitude above the IC_{50} of 6 nM (K_i of 600 pM). In contrast, no TPPU was detected in the control mice (n=4). **B.** Whole blood was collected via cardiac puncture, and plasma was isolated and analyzed for changes in EET to DHET ratio. Chronic TPPU treatment has significantly increased the EET/DHET ratio (n=4, p=0.036). *P < 0.05 and **P<0.01, as determined by the Two-tailed t-test. Bars represent ±SEM.

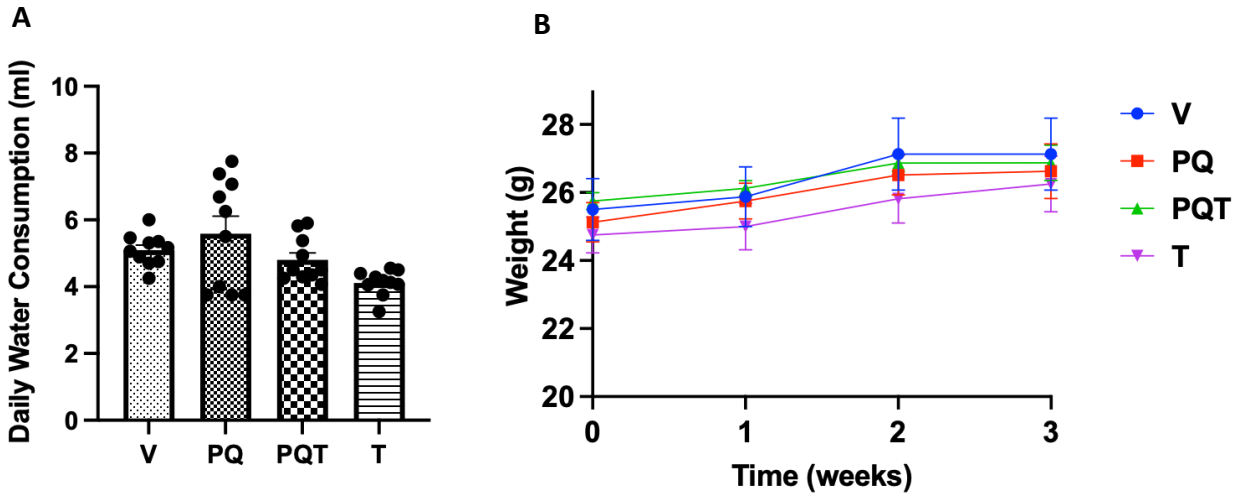

Figure S6. Measures of **A.** the estimated amount of water consumed per mouse per day, calculated from the total decrease in water volume by cage, and **B.** the mean weight of the mice in each treatment group (n=8). Mice in the TPPU group were slightly lighter than vehicle control mice ($p=0.009$) and drank less water than the vehicle control group ($p=0.016$), likely due to their lower initial weight. Abbreviations: V = vehicle, PQ = paraquat, PQT = paraquat and TPPU, T = TPPU only control. Results are expressed as mean ± SEM. Statistical significance was determined by the Kruskal-Wallis test or repeated measures one-way ANOVA followed by Dunnett's post-hoc test.

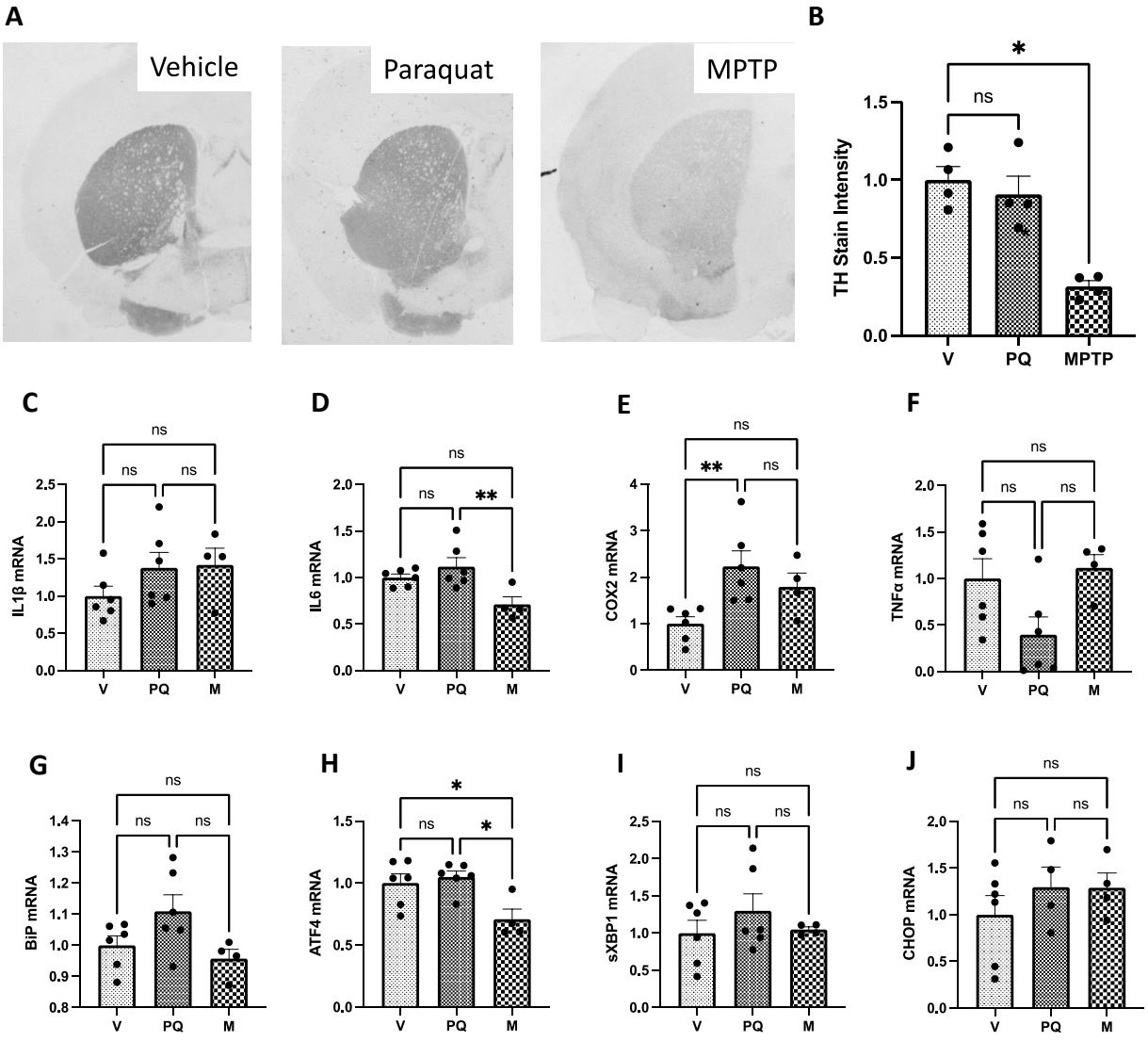

Figure S7. Immunohistochemical analysis of striatal expression of tyrosine hydroxylase and RT-qPCR of striatal mRNA. Mice were treated with MPTP (10 mg/kg/day, *i.p.*) in sterile PBS for five days and were euthanized five days post final injection to compare its effects to those of PQ treatment. (**A-B**) While MPTP resulted in a significant decrease of tyrosine hydroxylase indicative of dopaminergic neural loss (n=4, $p<0.001$), it did not significantly increase any inflammatory or ER stress markers. (**C-J**) MPTP resulted in reduced GAPDH normalized mRNA expressions of IL-6 ($p=0.005$) and ATF4 ($p=0.014$) compared to PQ treatment and reduced ATF4 ($p=0.039$) compared to vehicle control. Vehicle and PQ data are equal to those presented in Figure 3. MPTP treatment was conducted simultaneously with the PQ treatment. Abbreviations: V = vehicle, PQ = paraquat, M = MPTP. Results are expressed as mean ± SEM. *$P < 0.05$ as determined by one-way ANOVA or Kruskal-Wallis test followed by Dunnett's post-hoc test.

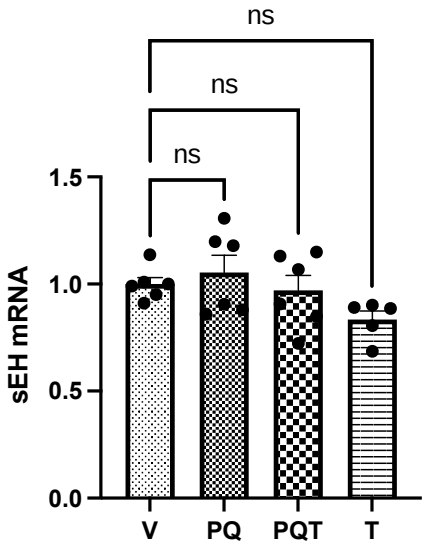

Figure S8. Effect of PQ and TPPU on striatal GAPDH normalized sEH mRNA expressions. No significant difference in the level of sEH gene expression was detected between vehicle control and PQ- or TPPU-treated mice when assessed via post hoc test. Results are expressed as mean ± SEM. RT-qPCR, data represent 5 or 6 biological replicates. Abbreviations: V = vehicle, PQ = paraquat, PQT = paraquat, TPPU, T = TPPU only control. *P < 0.05 and **P<0.01, as determined by one-way ANOVA followed by Dunnett's post-hoc test.