Supporting Information

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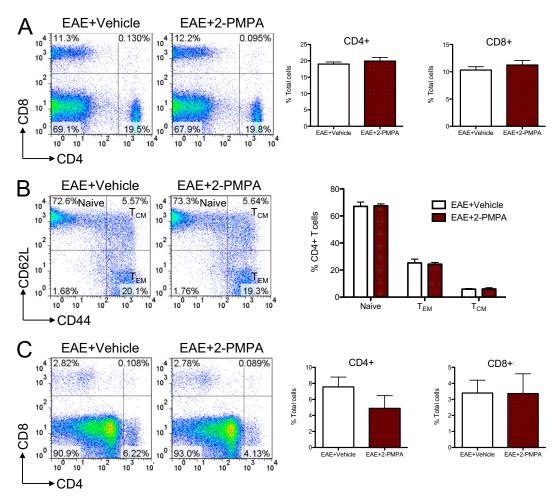


Fig. S1. Effect of 2-PMPA on immune cell populations. Daily administration of 2-PMPA in EAE mice does not affect splenic or CNS lymphocyte populations 28 d postimmunization. (A) Splenocytes isolated from 2-PMPA-treated and control mice on day 28 postimmunization were stained directly ex vivo for CD4 and CD8. Cells were gated on live lymphocytes, and percentages of CD4* (Left) and CD8* (Right) T lymphocytes were determined. (B) CD4* T cells from A were stained for CD44 and CD26L. Percent of each subtype, T effector memory (T_{EM} CD44^{hi} CD62L^{lo}), T central memory (T_{CM} CD44^{hi} CD62L^{hi}), and T naïve (CD44^{lo} CD62L^{hi}), are shown in the graph. (C) Mononuclear cells isolated from brains of mice used in A were stained directly ex vivo for CD4 and CD8. Percentages of infiltrating CD4* (Left) and CD8* (Right) T lymphocytes were determined. Representative FACS plots are shown. Statistical significance was determined with P < 0.05. n = 4 mice per group.

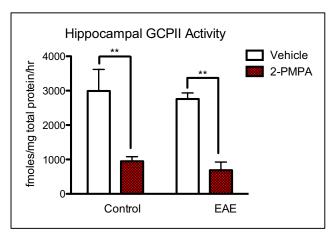


Fig. S2. GCPII activity is equivalent in control and EAE mice. Control and EAE mice have similar levels of hippocampal GCPII activity. Daily administration of 2-PMPA for 15 d decreases the level of GCPII activity compared with vehicle-treated counterparts by \sim 68 and 75% in control and EAE mice, respectively. Significant difference is shown from control + vehicle or EAE + vehicle at P < 0.01 (**). n = 3 mice per group.

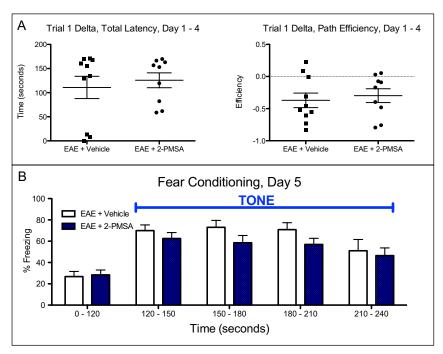


Fig. S3. Effect of 2-PMSA on cognition in EAE mice. Daily administration of 2-PMSA, an inactive analog of 2-PMPA, does not affect Barnes maze performance as indicated by total latency delta (A, day 1 total latency – day 4 total latency) and path efficiency delta (day 1 path efficiency – day 4 path efficiency) compared with vehicle-treated controls. Daily administration of 2-PMSA does not affect fear conditioning performance (B) compared with vehicle-treated controls. A = 10 mice per group. Statistical significance was determined with A < 0.05. A = 9–10 mice per group.

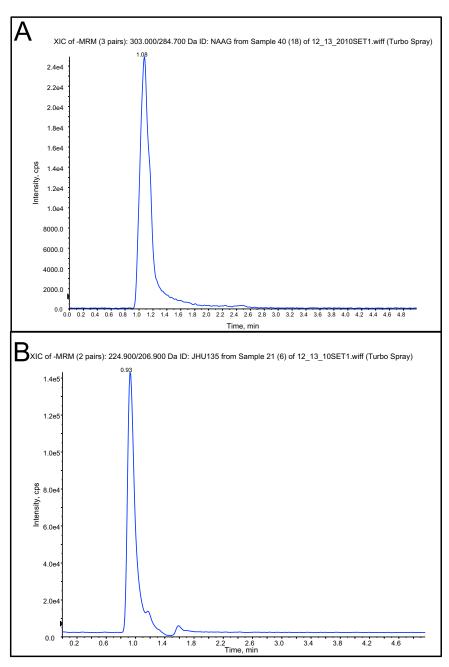


Fig. S4. Sample spectra of mouse brain NAAG (A) and 2-PMPA (B). Chromatographic separation for NAAG and NAA in brain tissue homogenates was achieved using Waters Atlantis column and mobile phase delivered isocratically (acetonitrile: 2 mM ammonium acetate; 35:65, vol/vol; containing 0.1% formic acid). NAA, NAAG, and the internal standard were monitored by tandem mass spectrometry in electrospray negative ionization mode. Linear calibration curves were generated over the range of 5–5,000 ng/mL and tissue samples quantified as μg/g. For determination of 2-PMPA, separations were achieved on a Waters XTerra MS C18 column, using MeOH and 2 mM N, N-dimethylhexylamine buffer (50:50, vol/vol at pH 8.5) delivered isocratically. 2-PMPA and the internal standard were monitored by tandem mass spectrometry with electrospray negative ionization mode. Linear calibration curves were generated over the 1–100 ng/mL range and converted to μg/g.