Putative Microcircuit-Level Substrates for Attention Are Disrupted in Mouse Models of Autism

Supplemental Information

Supplemental Methods & Materials

Matching Levels of Activity Between Datasets

To reduce activity in one group of datasets in order to match levels of activity in another group of datasets (c.f. Fig. S1C,D; Fig. S2C), we randomly deleted epochs of activity until the % time active in a given dataset matched the mean level of activity in the second group. For example, to generate Fig. S2C, we deleted randomly selected epochs of activity from saline-exposed datasets until they reached a mean level of activity of 1.4%, corresponding to the mean level of activity in VPA-exposed datasets. All data shown represents an average based on 5 instantiations of this epoch removal algorithm.

The upsampling procedure performed in Figure S3 was achieved by overlaying multiple datasets to produce a single, virtually upsampled dataset. For example, we combined 2, 3, or 4 saline-exposed datasets or 3, 5, or 7 VPA-exposed datasets to achieve a higher-than-normal density of events, as indicated in the Fig. S3 legend. The correlation distributions represent the means, computed over all possible combinations of N datasets drawn from the total number of experiments for each condition. The datasets were combined by simply computing the union of the two datasets to produce a new dataset with a number of virtual neurons equal to the lesser of the numbers of cells in the two experiments, e.g. overlaying two VPA datasets, with 82 neurons and 84 neurons, yields a combined dataset with 82 virtual neurons. In the combined dataset, virtual neuron i is active at time t if neuron i is active at time t in either of the original datasets.

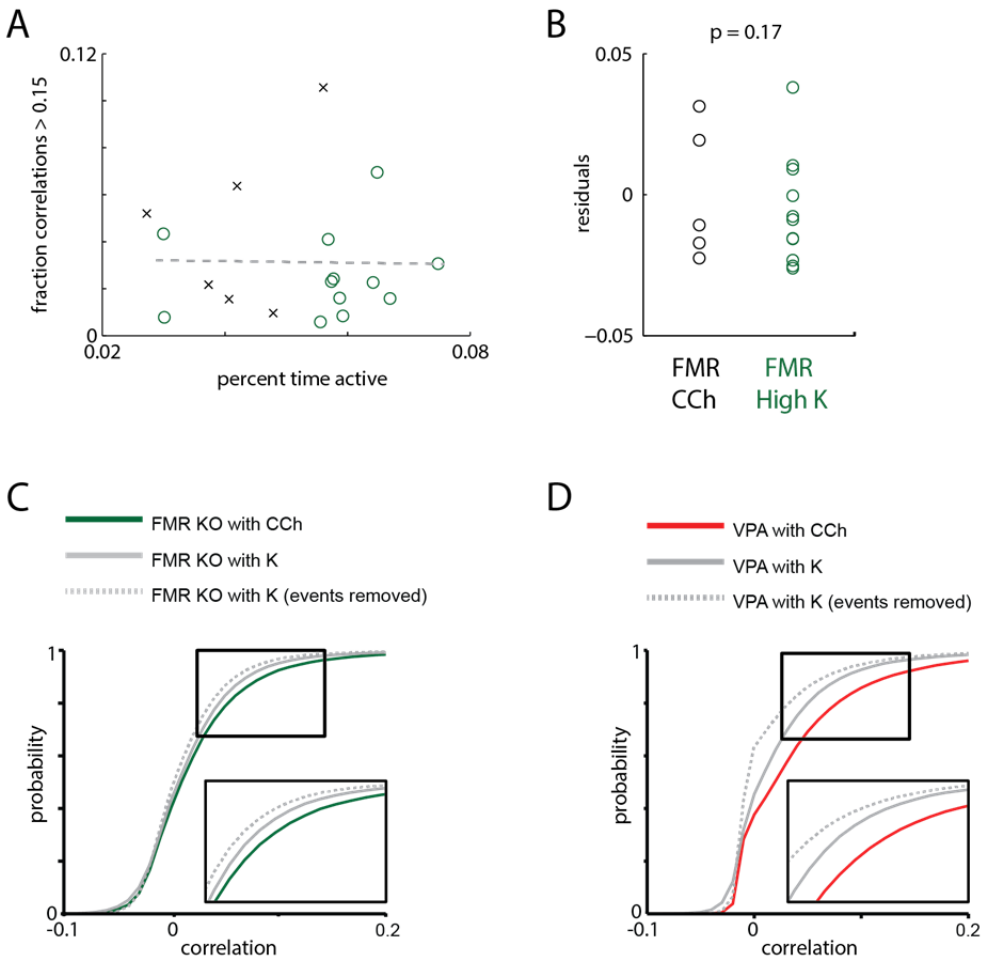

Figure S1. Differences in activity levels do not explain the inability of cholinergic modulation to decorrelate activity in autism models. (A) Scatterplot showing the fraction of strong correlations as a function of the mean % time active in *FMR1* KO mice in carbachol (black) or high K^+ ACSF (green). The gray dashed line represents a linear fit of a linear model to all points (carbachol + high K^+ ACSF). **(B)** Residual values for the fraction of strong correlations in carbachol (black) and high K^+ ACSF (green), i.e. the difference between the actual values and the number expected based on a linear relationship between the level of activity and the prevalence of strong correlations. Even after accounting a possible relationship between activity and correlations, there is no carbachol-induced decorrelation in *FMR1* KO circuits. **(C)** Cumulative distribution plot for correlations recorded from *FMR1* KO mice in carbachol (green line) or high K^+ ACSF (gray line). The cumulative distribution plot is also shown for datasets recorded in high K^+ ACSF that have had epochs of activity removed to match activity levels observed in carbachol (dotted gray line). These plots show that removing epochs of activity tends to *decrease*, rather than increase, correlations, suggesting that abnormally high

correlations in *FMR1* KO mice in carbachol are not an artifact of decreased activity levels. **(D)** An analysis based on linear regression and residual values was not possible for the VPA datasets, because activity levels were largely non-overlapping for the carbachol and high K^+ conditions, making it impossible for linear regression to dissociate activity-driven differences in correlations from those driven by condition (carbachol vs. high K^+). Instead, we again removed epochs of activity from VPA-exposed datasets in high K^+ ACSF to match the levels of activity in carbachol. Cumulative distribution plots are shown for correlations recorded from VPA-exposed mice in carbachol (red line), high K^+ ACSF (gray line), and high K^+ datasets that have had epochs of activity removed to match activity levels observed in carbachol (dotted gray line). Our finding that VPA-exposed circuits exhibit a carbachol-induced *increase* (rather than decrease) in strong correlations cannot be explained simply by the fact that activity levels are lower in carbachol than in high K^+, since lower activity levels tend to decrease, rather than increase, correlations.

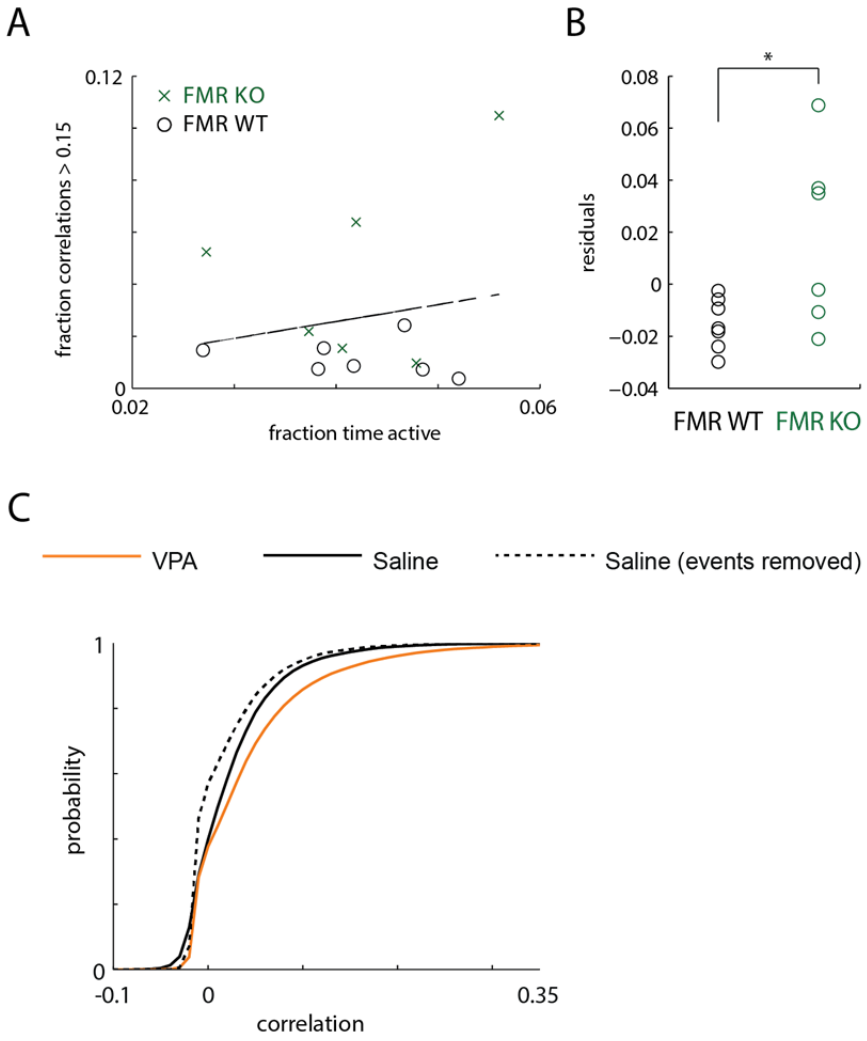

Figure S2. The abnormally increased correlations observed in two models, relative to controls, in carbachol, are not an artifact of differences in activity levels. (A) Scatterplot showing the fraction of strong correlations as a function of the mean % time active for *FMR1* WT mice in carbachol (black O symbols) or *FMR1* KO mice in carbachol (green X symbols). The gray dashed line represents a linear fit of all points. **(B)** Mean activity levels were almost identical for *FMR1* WT (black) and *FMR1* KO (green) datasets in carbachol. Nevertheless, we analyzed residual values based on the linear regression shown in (A) to confirm that the increased correlations in *FMR1* KO mice in carbachol were not an artifact of differences in activity. We plotted residual values for the fraction of strong correlations in *FMR1* WT (black) and *FMR1* KO (green) datasets in carbachol, i.e. the differences between the actual values and the values expected based on a linear relationship between the fraction of strong correlations and mean % time active. Even after accounting for a possible relationship between correlations and activity, residual correlations in *FMR1* KO mice are still significantly greater than those in

FMR1 WT mice. **(C)** Again, an analysis based on linear regression and residual values was not possible for the VPA-exposed datasets and saline-exposed controls, because activity levels were largely non-overlapping for these two conditions, making it impossible for linear regression to dissociate activity-driven differences in correlations from those driven by condition (VPA-exposed vs. saline-exposed). Instead, we created surrogate datasets to artificially match levels of activity between these two conditions by either removing epochs of activity (here), or by overlaying multiple datasets to increase levels of activity (in Fig. S3). Here we have plotted the cumulative distributions for correlations recorded in carbachol from VPA-exposed (red line) or saline-exposed mice (gray line). Reducing activity in control datasets, by removing epochs of activity to match the activity levels observed in VPA-exposed datasets (dotted gray line), tends to decrease correlations and exacerbate the difference between these two populations. Thus, the increased strong correlations in VPA-exposed datasets, compared to controls, does not appear to be an artifact of differences in activity levels. * $p < .01$ by Wilcoxon rank sum test.

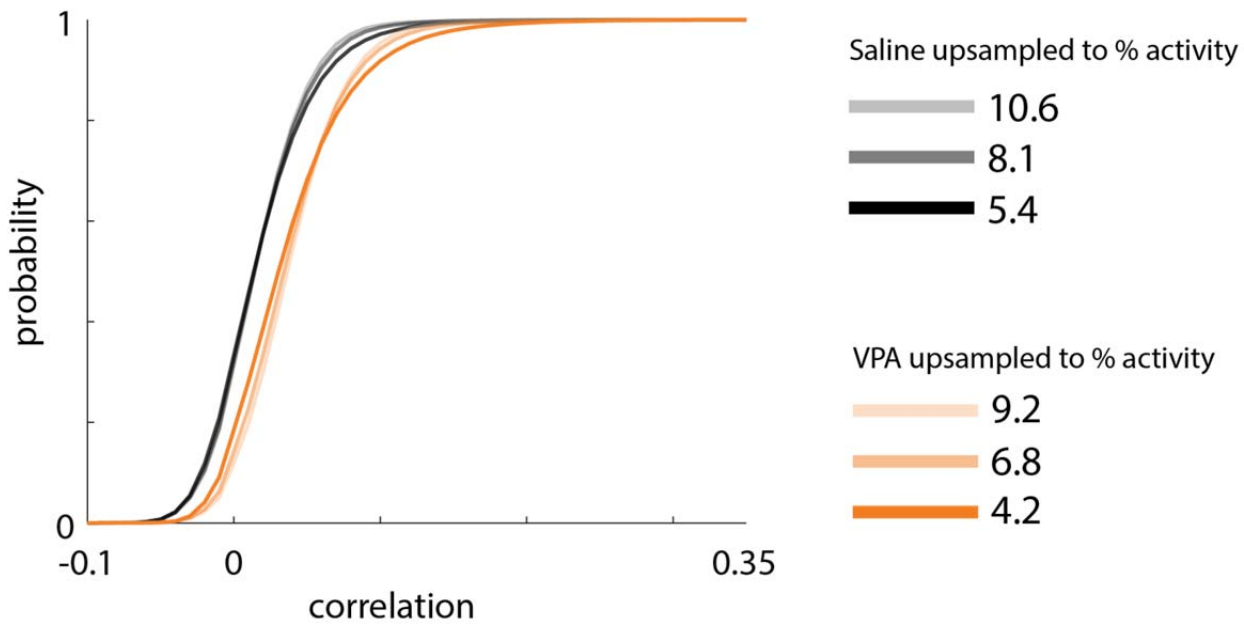

Figure S3. VPA-exposed datasets continue to exhibit an increase in strong correlations, even after matching the higher levels of activity in saline-exposed datasets. Generating surrogate datasets by overlying multiple actual datasets to artificially increase levels of activity (described in the Methods), shows that regardless of the level of activity, correlations are consistently increased in VPA-exposed datasets in carbachol (orange) compared to saline-exposed controls in carbachol (black). The legend shows the mean % time active for each surrogate dataset generated by overlaying multiple VPA-exposed datasets or saline-exposed controls. This provides additional evidence against the idea that the increased correlations observed in VPA-exposed datasets are simply an artifact of differences in activity levels.