Abnormal Activity-Dependent Brain Lactate and Glutamate+Glutamine Responses in Panic Disorder

Supplemental Information

Voxel Segmentation Procedures

Immediately after collection of all magnetic resonance spectroscopy (MRS) data, a high-resolution, 3D-SPGR image was acquired using a quadrature head coil. Scanning parameters were TE/TR = 7/34; flip angle = 35; bandwidth = 15.63; resolution = 0.86 mm isotropic in-plane resolution with 1.6 mm slice thickness; coronal slices = 124. The 3D-SPGR image was acquired in 17 of the 21 panic disorder (PD) patients and in 11 of the 12 control subjects.

The MRS voxel was mapped onto the SPGR image using FSL tools (1). First, the transformation of the T2 localizer scan into the SPGR space was calculated using the FMRIB Linear Image Registration Tool and the Brain Extraction Tool (2,3). Second, the location information for the MRS voxel was read from the spectroscopy file header, and a voxel mask was created on the T2 localizer image used to prescribe the voxel (Tanase, C., unpublished). Third, the voxel mask was mapped onto the SPGR image using the previously calculated T2 to SPGR transformation. Finally, using the FSL Segmentation Tool (4), the gray matter, white matter and cerebrospinal fluid partial volume maps were generated and the segmentation of the voxel was evaluated using direct summation of the partial volume maps over the MRS voxel mask in SPGR space.

Current and Past Comorbid Diagnoses

Among the thirteen remitted patients, past diagnoses included PD with agoraphobia in ten, alcohol abuse in five (median interval 21 years prior), major depression (MD) in three, social phobia (SP) in two, and generalized anxiety disorder (GAD), posttraumatic stress disorder, and anorexia nervosa in one each. Among the ten patients with symptomatic, primary PD, current diagnoses included PD with agoraphobia in nine, GAD in three and SP in two. Past diagnoses included MD in five and drug dependence in one (23 years prior). Of the thirteen healthy control subjects, two had a past history of alcohol abuse (mean interval 14 years prior).

Example Spectra Showing LCModel Fit

Figure S1 shows two examples of the quality of the LCModel fit for spectra acquired using the CHESS pulse sequence over a single 5 minute period of visual stimulation.

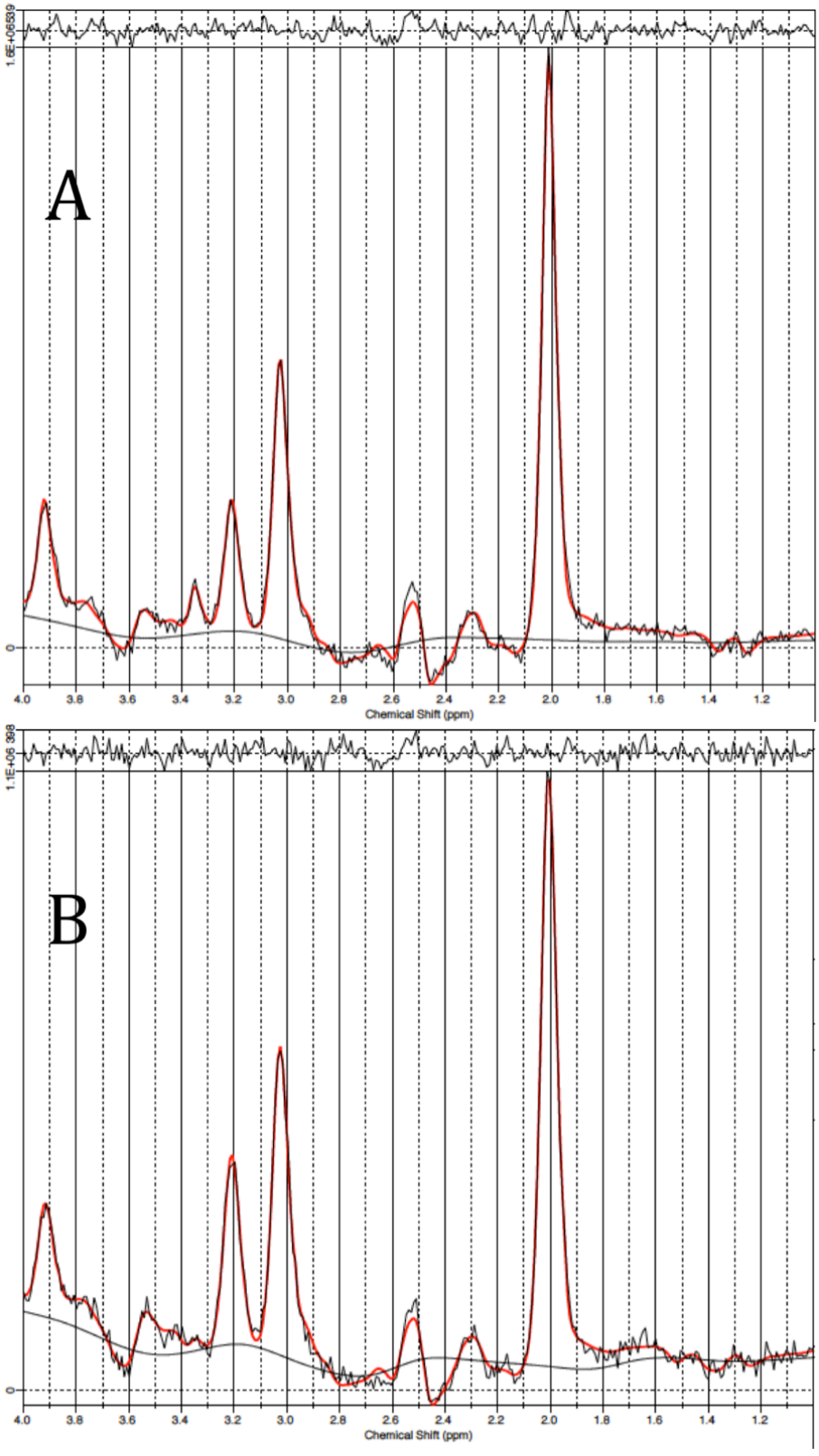

Figure S1. An example spectrum and its LCModel fit is shown for a remitted PD patient (panel **A**) and for a control subject (panel **B**). The large area in the center of each panel shows three

tracings: 1) the spectral data as acquired (black line); 2) the LCModel fit of the acquired spectral data (red line); and the LCModel baseline estimation (black line along the baseline of the spectral data). The small area above the spectral data shows the residual signal (black line), which illustrates how much the acquired spectral data deviates from the fit. Each spectrum was obtained from the interleaved CHESS frames acquired over a single 5-minute period of visual stimulation (see Methods). CHESS, chemical-shift selective saturation.

Temporal Correlation Between Changes in Lactate and Changes in Glx

The temporal association between activity-dependent changes in lactate/NAA and glx/NAA was assessed by correlating the percent change values of the two metabolites over the four time periods (VS1, VS2, RC1 and RC2) within each subject. The correlation coefficient (Pearson's r) for each subject was converted to a z-value, based on the probability distribution of r. The control subjects had significantly higher z-values than the PD patients. Figure S2 shows the simple linear regression lines calculated between change in lactate on the Y axis and change in glx on the X axis over the four time periods for PD patients and control subjects.

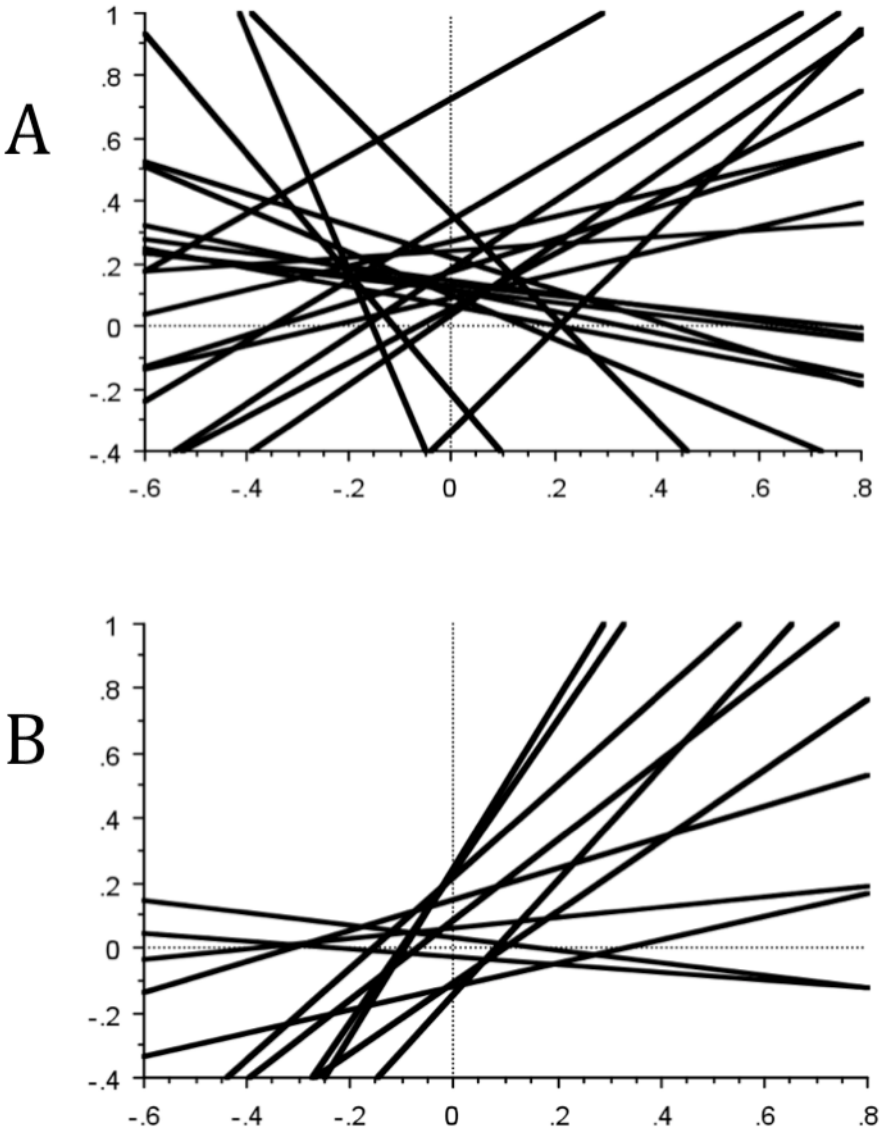

Figure S2. Each line illustrates the slope of the regression between changes in lactate over time (Y axis) and changes in glx over time (X axis) for each of the 20 PD patients (**A**) and each of the 11 control subjects (**B**) (outliers excluded). In the control subjects, all but two subjects show a positive slope. In the PD patients, there is no consistent relationship between change in lactate and change in glx over time. glx, glutamate + glutamine.

Supplemental References

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2. Jenkinson M, Smith SM (2001): A global optimisation method for robust affine registration of brain images. *Med Image Anal* 5:143-156.

3. Smith SM (2002): Fast robust automated brain extraction. *Hum Brain Mapp* 17:143-55.

4. Zhang Y, Brady M, Smith S (2001): Segmentation of brain MR images through a hidden Markov random field model and the expectation-maximization algorithm. *IEEE Trans Med Imaging* 20:45-57.