Inhibitory Functioning in Fear Extinction: GABA and BOLD Responses

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Introduction. Impaired fear extinction plays a critical role in the vulnerability for anxiety disorders. The GABAergic system is thought to play a key role in the regulation of fear and its dysregulation may contribute to the development of pathological anxiety. Numerous neuroimaging studies have identified the dorsal anterior cingulate cortex (dACC) as one of the main brain regions involved in fear expression[1]. The objective of this study was to investigate the impact of individual differences in dACC GABA concentrations on the neural basis of fear acquisition and extinction in healthy individuals. We performed a multimodal imaging study combining GABA magnetic resonance spectroscopy (MRS) with functional MRI.

Materials & Methods. 60 healthy young males participated in a fear conditioning and extinction paradigm during a functional MRI session. Scanning was performed on a Philips Achieva 3.0T TX using a 32-channel receiver head-coil (Philips Achieva 3.0T TX, Philips Healthcare, The Netherlands). The MRI protocol included a MPRAGE anatomical scan to plan the dACC MRS voxel. Subsequently, GABA MRS spectra in the dACC were acquired using a MEGA-PRESS sequence[2] with the following parameters: TR/TE=2000/73, 384 averages, voxel size=40x20x20mm³, T_{acq}=12:48min. Following MRS acquisition, subjects underwent a fear conditioning and extinction task during a BOLD fMRI sequence (TR/TR=2000/27.63, GE-EPI read out, FOV=240x240mm², voxel size=3x3x3mm³, 37 slices). The task involved the presentation of two differently colored squares as conditioned stimuli. During the conditioning phase, one of the conditioned stimuli (CS+) was paired with an unconditioned stimulus (US: mild electric shock) with a reinforcement rate of 33% while the second conditioned stimulus (CS-) was never paired with a US. In the subsequent extinction phase the conditioned stimuli were presented but no shocks were administered. Both conditioning and extinction training consisted of 36 trials.

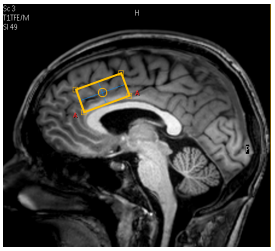

Figure 1: dACC GABA MRS voxel placement (40x20x20mm³).

GABA MRS spectra and fMRI data of 60 subjects were analyzed using AMARES in jMRUI[3] and SPM8 (Wellcome Trust Center for Neuroimaging, www.fil.ion.ucl.ac.uk/spm), respectively. After a manual phase adjustment procedure, MRS spectra were analyzed for GABA and NAA. For GABA quantification, a single Gaussian was fitted onto the MRS difference spectra at 3ppm. The NAA peak was fitted at 2.01ppm using a single Lorentzian and was used as the reference metabolite to quantify GABA (GABA/NAA ratio in institutional units [i.u.]). Forty-four complete data sets were included in the final analysis (data sets of sixteen participants were excluded based on either insufficient SNR of MRS spectra or incomplete fMRI data sets).

Results. GABA levels obtained from the dACC were quantified relative to NAA concentrations and ranged from 0.037 i.u. to 0.333 i.u. (mean=0.176 i.u.; SD=0.065i.u.). To verify successful conditioning we contrasted BOLD signal changes of the CS+ and CS- during the conditioning phase which revealed a strong increase in BOLD activity in the anterior cingulated cortex (Fig. 2; cluster-level: $MNI_{x,y,z}$=(6,4,42)), P_{FWE}<0.001). In order to obtain a neural index of individual differences in extinction learning, the extinction trials were split up into early (18 trials) and late (18 trials) phase extinction training. As expected, the dACC showed lower activity during late compared to early extinction (Fig. 3; $MNI_{x,y,z}$=(-4,2,42)), P_{FWE}<0.001). Subsequently, we extracted fMRI extinction data from the MRS voxel region of interest. A correlation analysis showed that individuals with low dACC GABA concentrations demonstrated reduced extinction to aversive stimuli at trend level (Fig. 4; r=0.3, p=0.055).

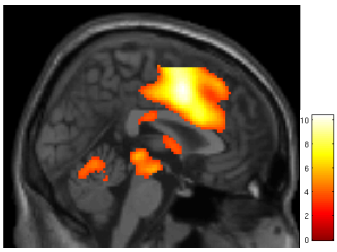

Figure 2: Increased group BOLD activity in response to CS+ presentation relative to CS- presentation during the conditioning phase (threshold t>3.1, FWE-corrected)

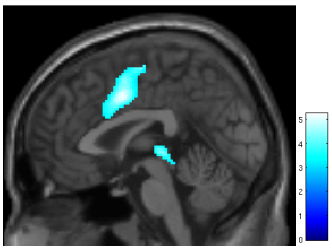

Figure 3: Activity changes in response to the CS+ during extinction training: group BOLD activity decrease from early phase to late phase extinction training (threshold t>3.1, FWE-corrected).

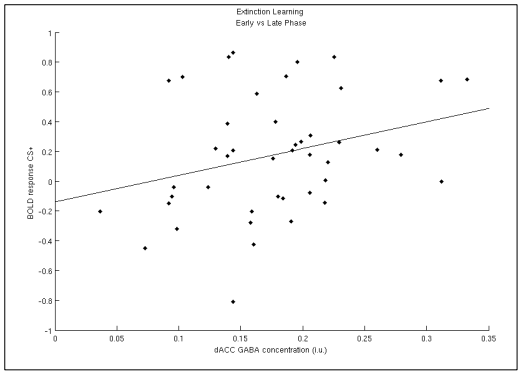

Figure 4: Correlation of dACC GABA concentrations and BOLD signal changes to the CS+ from the early to the late phase of extinction training showing a positive trend (r=0.3, p=0.055).

Discussion and Conclusion. We hypothesized that individuals with low dACC GABA concentrations would show reduced fear extinction compared to individuals with high GABA levels. Preliminary results show a trend towards this: individuals with low GABA levels displayed smaller differences in BOLD responses between early and late phase of extinction when presented with the CS+ compared to participants with high GABA levels. This might indicate that individuals with low GABA levels at rest perform worse in learning a new safety association for the previously aversive cue. The present findings provide information on the importance of metabolite differences in neuroimaging research and their effects on cognitive functioning. Furthermore, these results might be a first step towards a new insight into predicting individual vulnerability and treatment response in anxiety disorders.

References:

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