Graph theoretical Network Analysis of Pain Processing: Pain is more than Sensation

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Introduction: Modern neuroimaging techniques, such as functional MRI, have allowed us to investigate the neural basis of pain perception and have shown that nociceptive stimuli commonly elicit activity within a very wide array of subcortical and cortical brain structures [1,2]. These structures and their interactions are usually referred to as "Pain matrix" and are considered to be specific for the processing of nociceptive stimuli [3,4]. Recently this widely accepted theory is queried: the so called "Pain matrix" may not be specific for nociceptive stimulation but instead may be involved in processing of salient input of all sensory systems independent of their modality [5]. Both hypotheses rely mainly on the description of the evoked activity of the individual brain structures. Recently investigations of the information flow between such regions, i.e. interregional connectivity, came into focus, providing additional information helping to better understand this cognitive processing [6]. Therefore, we investigated changes in the interregional connectivity of neutral versus painful (thermal) stimuli in a mouse model.

Material and Methods: Experiments were performed on 10 male wild-type mice (10-14 weeks old). Animals were anesthetized with 1.2% isoflurane in medical air. Body temperature was maintained at 37°C by warm water circulating throughout the holding cradle. *Image data acquisition*: fMRI experiments were performed with a 4.7 T/40 cm horizontal bore actively shielded magnet BioSpec (BRUKER, Germany). Gradient system (200 mT/m) and whole-body birdcage resonator enabled homogenous excitation. An actively RF-decoupled quadrature head coil was used as a receiver coil. Functional BOLD MRI scans were performed using a T2*-weighted single-shot gradient echo EPI sequence (22 axial slices, 64 x 64 matrix, TR= 2000 ms, k-space averaging of 2, TEef= 24.4 ms, field of view 15 x 15 mm, in-plane spatial resolution 234 x 234 µm, slice thickness 500 µm). *Experimental protocol*: a set of single thermal stimuli (40°,45°,50° and 55° ± 1°C, plateau for 5 s, ramp 15 s) was applied three times in 3 min 25 sec intervals by a Peltier element on the right hindpaw. *Data processing*: after motion correction, general linear modeling analysis with separate predictors for each comparisons, data were FDR thresholded (q<0.05), and different groups of activated voxels were labeled as belonging to 150 pain related brain structures based on the mouse atlas from Paxinos [7].*Connectivity analysis*: The average time courses of the activated voxels of each brain structure were corrected for global signal fluctuations by linear regression using the global mean as regressor. Residual time courses were crosscorrelated individually for each stimulus and animal and the resulting 150x150 crosscorrelation matrices were averaged over animals. The mean crosscorrelation matrices were used to create a stimulus specific network with correlation coefficients as weights and brainstructures as nodes. Conventional graph theoretical analysis was performed [8]. Modules were identified using the Blondel community detection [9] and hubs using the Hyperlink-Induced Topic Search (HITS) by Jon Kleinberg [10].

Results: All networks show high modularity, which is a typical feature of non-random small-world networks. Two modules are stable over all stimulation temperatures. The first one contains brain structures of the sensory input, hippocampal structures and the PAG (SI-module). The second one contains basal ganglia, structures of the association and motor cortex, and septum (BA-module) (Fig. 1). Other structures like thalamus, somatosensory cortex and amygdala change their module affiliation especially at the transition from the non painful stimulus (45°C) to the first painful stimulus at 50°C. In the network of the 45°C non-painful stimulation the areas with the highest degree and hub functionality are part of the SI-module. The very same structures seem to have no important function in the topology of both painful stimuli. During pain processing areas of the BA-module gain important hub function. The node specific differences in degree and average BOLD amplitude between painful and non-painful stimulation showed a significant positive linear correlation (Fig. 2). Structures of the BA-module show an increasing degree and response amplitude whereas the structures of the SI-module loose connections (decreasing degree) along with a small amplitude increase or even decrease.

Disussion: We could show that the analysis of interregional connectivity provides additional information concerning the cognitive procession of nociceptive stimuli. Descriptive parameters such as size of activated area and activity show a linear increase over all stimulation temperatures [11] and thus are not sufficient to differentiate between pure somatosensory and nociceptive stimuli. However, stimulus specific interregional connectivity networks demonstrate the rebuilding of connections between brain structures during nociceptive processing. This indicates that processing of painful stimuli differ from the processing of other salient ones in their interregional connectivity. In conclusion, stimulus specific network analysis gives us a deeper insight into the functionality of the brain and thus may help us to understand the emergence and processing of pain.

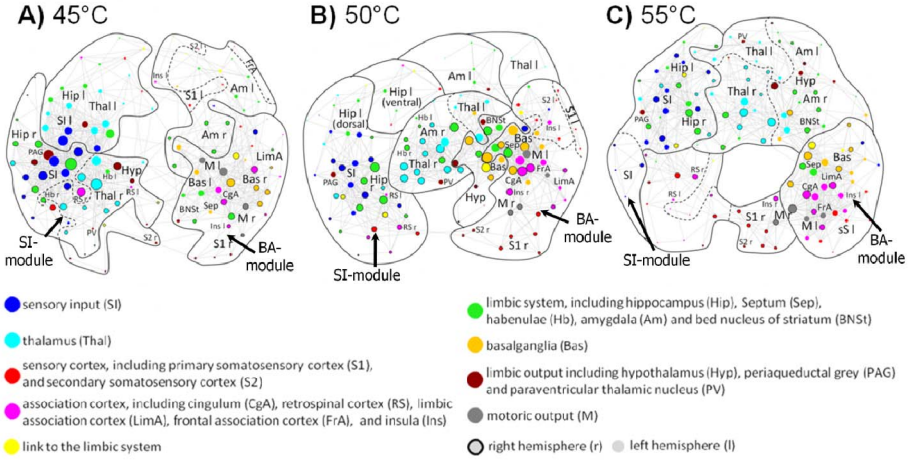

Fig. 1: Stimulus specific networks (150 nodes, top 750 connections, isolated nodes removed). Node size encodes the node's degree. Outlines encase the modules identified by Blondel's community algorithm [9] (dashed lines communities on level 0, solid lines on level 1).

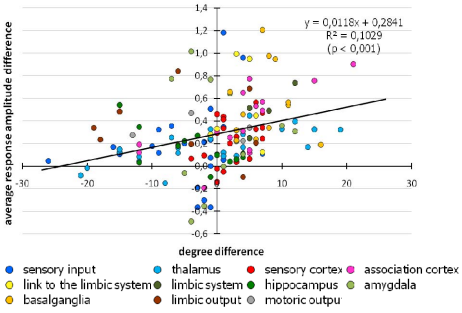

Fig. 2: Linear correlation of node specific degree difference and node specific difference of the average BOLD amplitude between painful and non-painful stimulation (50°C – 45°C).

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