Repeated Resting State fMRI During Dose-Controlled Morphine and Alcohol Infusion Reveals Localized and Drug Specific Changes in Functional Brain Connectivity

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Introduction: Low frequency fluctuations in the BOLD signal in the brain 'at rest' elicit topological patterns of functional connectivity [1]. Growing evidence for changes in functional connectivity in CNS diseases and consciousness states provides incentive that resting state fMRI (RS-fMRI) be explored in CNS drug research. Applicability of task-fMRI in drug research has been previously illustrated [2]. However, *a priori* expectations of effects in regions of interest or specific behavioral responses limit the scope of research models. More importantly, interindividual variability in task-training and performance are subjective and can hardly be modeled. Thus, RS-fMRI may be more advantageous since the input (e.g. drug dose or concentration) and the pharmacokinetic effects can be objectively modeled in experimental designs. In this study, we use repeated RS-fMRI series in a within-subject placebo-controlled pharmacological study to investigate whether controlled doses of morphine and alcohol lead to regionally specific changes in CNS connectivity at rest.

Methods: Twelve healthy young men participated in a double blind, placebo controlled repeated measures study consisting of three examination sessions. In each procedurally identical session, drug compounds were administered intravenously. Infusion protocols were based on validated pharmacokinetic models to minimize between- and within subject variations and ascertain controlled levels of plasma drug concentration across time [3]. Seven BOLD RS-fMRI data sets were acquired:1 sample before infusion, 3 samples at 30 minutes intervals after the drug had reached the desired concentration (0.6 Kg/L for ethanol and 80 nmol/L for morphine), and 3 more samples (30 minutes apart) after stopping the drug infusion. Monitoring and registration of physiological signal (heart and respiration) was conducted during fMRI acquisition. Drug concentrations were measured between scanning intervals from breath alcohol and plasma morphine. Visual analogue scales were repeatedly administered to measure subjective feelings of mood (e.g. highness, alertness and calm) and physical wellbeing (nausea). In total, 252 data sets (21 sets per person) were collected. Using FSL, we applied the dual regression method [4] to describe voxel-wise functional connectivity to eight pre-defined CNS networks (i.e. z-scores of fitting fluctuations in each voxel to a representative time course of fluctuations in template networks). Reliability, consistency and functional relevance of these resting networks has been illustrated previously [1]. A general linear model was used to test effects of drug by time interactions on brain connectivity. Statistical significance (cluster corrected) was set at $p < 0.01$.

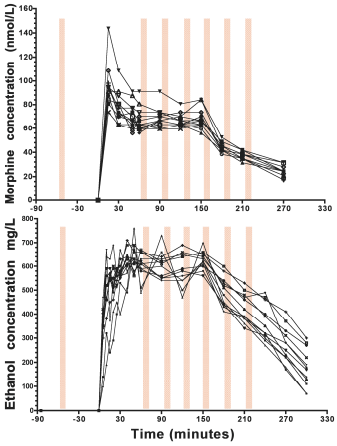

Figure 1: Individual variations in drug concentration for morphine (top) and ethanol (bottom). Red bars show when RS-FMRI was acquired.

Results: Stable drug concentration between minutes 60-150 was achieved (morphine, $F(3,11)=0.48$, $p > 0.69$; alcohol $F(3,11)= 1.24$; $p>0.30$). Average morphine and alcohol levels were in the target range of experimental design (Figure1). Compared to placebo and ethanol, morphine increased calmness, increased nauseous sensation and lowered the respiration and heart rates ($p<0.05$). Compared to placebo and morphine, alcohol increased subjective feeling of drunkenness, and increased the heart rate ($p<0.05$); but had no significant effect on other mood scales, or on the respiration rate. Ethanol and morphine had distinguishable effects on functional brain connectivity (Figure 2). Compared to placebo, effects of morphine over time were significant on connectivity of cingulate cortex, the hippocampus, cerebellum and prefrontal regions. By comparison, effects of ethanol compared to placebo were less extensive but detectable in the visual cortex, cerebellum and midbrain areas.

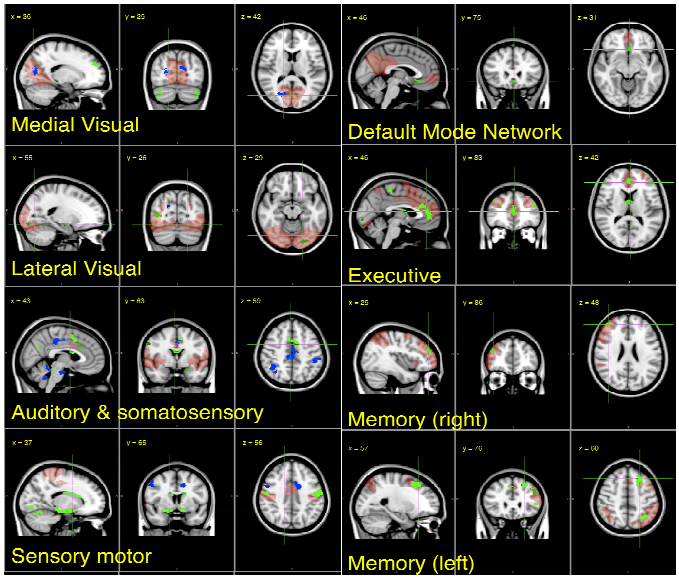

Figure 2: Template resting state networks (Red); changes in connectivity to template networks (Morphine Green; Ethanol Blue); p<0.01, corrected.

Conclusion: This is the first study to perform repeated RS-fMRI over several hours to examine changes in functional connectivity in response to controlled doses of different drug compounds in the same subject. We have detected drug-specific and focal changes in connectivity of regions in the pain pathways and visual attention stream, which are expected targets for morphine and alcohol actions, respectively. Our findings illustrate applicability of data-driven RS-fMRI in CNS drug research.

References:
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