Integration of neural networks activated by amphetamine in females with different estrogen levels: A functional imaging study in awake rats.

Dan Madularu[1], Jason R. Yee[2], William M. Kenkel[2], Kelsey A. Moore[2], Praveen Kulkarni[2], Waqqas M. Shams[1], Craig F. Ferris[2], and Wayne G. Brake[1]
[1]Concordia University, Montreal, QC, Canada, [2]Northeastern University, Boston, MA, United States

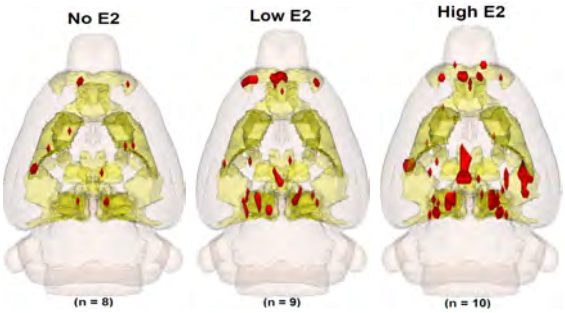
Target Audience: This experiment is targeted at clinicians and preclinical researchers alike.

Purpose: Previous studies demonstrate that schizophrenia symptomatology in women is dependent upon estrogen (E2) levels (1). Estrogen has beneficial properties when administered in conjunction with antipsychotics, and also alters dopamine neurotransmission (the target of most antipsychotic medications) in rats; suggesting a possible interaction between the two (2). The aim of the current study was to investigate this possible interaction using functional magnetic resonance imaging in awake, female rats.

Methods: Twenty-seven OVX, Sprague Dawley rats weighing 200-250 g were pair-housed in cages located in a 21^{0}C with a 12-h light-dark cycle (lights off at 19:00 h), with ad libitum access to food and water. Testing, injections, surgical procedures and imaging were performed during the dark phase of the diurnal cycle, in semi-dark conditions. Rats were divided into three groups, with respect to hormone replacement: no E2 (n = 8), constant low E2 (Low E2; n = 9) and constant low plus phasic high E2 (High E2; n = 10). The E2 (low and high) groups were implanted with sillastic capsules containing 17-β estradiol. High E2-replacement rats also received a subcutaneous injection of 17-β estradiol every second day (20 µg/kg dissolved in sesame seed oil) in a volume of 0.5 mL/kg body weight, providing an intermittent phasic high dose. Sensitization to the stimulant effects of repeated amphetamine (AMPH) has been widely used as a rodent model of some of the neurochemical and behavioural aspects of schizophrenia, and there are sex differences in its emergence (3).

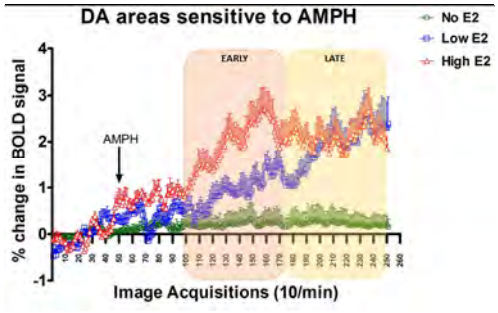
Rats were acclimated to the imaging system environment before being placed into the scanner itself. Animals were scanned at 300 MHz using a quadrature transmit/receive volume coil built into the rat head holder and restraining system for awake animal imaging (Animal Imaging Research, Holden, MA). The design of the coil provided complete coverage of the brain from olfactory bulbs to brain stem with excellent B1 field homogeneity. Rats were scanned using a Bruker 7T scanner and a 20-G/cm magnetic field gradient insert (ID = 12 cm) capable of a 120-µs rise time. At the beginning of each imaging session, a high-resolution anatomical data set was collected using the rapid acquisition with relaxation enhancement (RARE) pulse sequence (22 slice; 1.0 mm; field of vision [FOV] 3.0 cm; 256 × 256; repetition time [TR] 2.5 sec; echo time [TE] 12msec; NEX 2; 2 minute acquisition time). Functional images were acquired using a multi-slice Half Fourier Acquisition Single Shot Turbo Spin Echo (HASTE) pulse sequence. A single scanning session acquired 22 slices, 1.0 mm thick, every 6 s (FOV 3.0 cm, matrix size 96 x 96, NEX 1) repeated 250 times for a total time of 25 minutes. Each scanning session was continuous, starting with 50 baseline image acquisitions, then AMPH, followed by another 200 image acquisitions. Preprocessed functional files were then exported to Medical Image Visualization and Analysis (MIVA) for registration and segmentation. Images were aligned and registered to a 3D rat brain atlas (Ekam Solutions, Boston, MA, USA) which is segmented and labeled with 172 discrete anatomical regions. Scanning sessions consisted of 250 data acquisitions each, with a period of 6 seconds for each image, for a total time lapse of 1500 seconds or 25 minutes. The average signal intensity in each voxel of the first five minutes of baseline (acquisitions 1-50) was compared to acquisitions 100-175, and acquisitions 175-250 following AMPH. Volume of activation was compared across experimental groups using the nonparametric Kruskall-Wallis test statistic. In addition, two (early and late scan) separate repeated measures analyses of variance (ANOVA) were performed in order to assess the AMPH neural system time-course BOLD signal change in response to an AMPH challenge

Results. We found an overall effect of E2, whereby AMPH-sensitized, OVX females receiving phasic high E showed the highest BOLD activation in response to an AMPH challenge. In addition, it appears that the effects of E2 on BOLD activation are time-dependent, as shown by the difference observed during the early scan being minimized throughout the late scan.



Left: 17β Estradiol-dependent effect of amphetamine in the putative amphetamine neural circuit.

Right: BOLD signal change over time in response to amphetamine. Shown are average (+SEM) changes in BOLD activity of five components of the putative amphetamine neural circuit, averaged over animals.



Discussion. Taken together, our results suggest that 1) E2 interacts with dopamine in ROIs previously linked with schizophrenia such as the ventral tegmental area and medial prefrontal cortex, and 2) aspects of schizophrenia, as modelled in the AMPH-sensitized rat, are driven by more than a few select ROIs, such as the PFC and NAcc.

Conclusion. We show here that AMPH-induced, E2-dependent BOLD responses are rather global, involving entire pathways, as well as regulatory nuclei, such as the habenula. As a result, we propose that E2 has the potential of mediating most of the cognitive and behavioural effects seen in schizophrenia, as modelled via AMPH sensitization.

References.

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