Effect of chronic haloperidol treatment on the rat anterior cingulate cortex: linking neuroimaging findings with neuropathology

Anthony C. Vernon1, Winfred Chege1, William R. Crum1, Sridhar Natesan1, Michel Modo1, Jonathan D. Cooper2, Steven C.R. Williams2 and Shitij Kapur1

1Dept. Psychosis Studies, 2Dept. Neuroimaging, 3Dept. Neuroscience, Institute of Psychiatry, Kings College London, United Kingdom

Purpose:
- Neuropathological and neuroimaging studies suggest reduced gray matter volume of the cerebral cortex in schizophrenia, including the anterior cingulate cortex (ACC)1.
- It is unclear to what degree these changes are due to illness or antipsychotic medication intake. Notably, treatment with typical antipsychotics is associated with decreased ACC volume in first episode schizophrenia patients2.
- We have previously demonstrated that chronic haloperidol treatment at clinically relevant concentrations decreases the total neocortical volume of rats3,4. This provides a powerful model system to link neuroimaging changes induced by antipsychotics with those at the cellular level post-mortem to identify biological mechanisms.
- Using this rodent model, we first investigated the effect of chronic haloperidol treatment on brain volume using operator-independent deformation based morphometry (DBM) to localize drug-induced brain volume changes. The strongest DBM signal(s) in the cortex were then confirmed using manual segmentation and post-mortem histopathology to link volumetric changes identified by MR imaging with changes at the cellular level.
- We hypothesise that chronic haloperidol treatment is associated with a specific decrease in ACC volume in the rat neocortex.

Methods:
Male Sprague-Dawley rats were chronically administered either vehicle (n=8) or haloperidol (HAL; 2 mg/kg/day, n=7) using subcutaneously implanted osmotic minipumps for a total of 8 weeks5. MRI scans were acquired ex vivo as previously described6 and analyzed using whole brain and specific region-of-interest (cortex, hippocampus, striatum) DBM. The resulting statistical maps were corrected for multiple comparisons using the false discovery rate (FDR) procedure (p<0.05). To confirm DBM results, major cortical sub-field volumes were analyzed post-mortem from Nissl stained sections (1 μm thick) using unbiased stereology procedures (Cavalieri probe). Specific changes were further investigated by manual segmentation of ex vivo MRI images and stereological estimates of volume and neuronal number/density (Optical fractionator). These data were analyzed using two-tailed students t-test (SPSS v.20, IBM).

Conclusions
- DBM analysis suggests region-specific changes in the volume of the rat brain following chronic haloperidol treatment.
- Manual segmentation and post-mortem histopathology confirm that chronic haloperidol treatment induces a decrease in cortical volume, localized in the ACC. This was not explained by a change in neuron number, but is associated with increased neuronal density, suggesting a loss of neuropil.
- These data are consistent with findings from first episode patients treated with typical APD7,8. However, ACC abnormalities also predates psychosis onset9. Thus, typical antipsychotics may contribute to cortical volume changes, including the ACC, but cannot be the sole cause.
- These data highlight the utility of this pre-clinical model system to investigate the neurobiology underlying antipsychotic medication induced changes in brain structure.


Figure 1. DBM analysis pipeline

Table 1. Estimated Neuron number

<table>
<thead>
<tr>
<th>Cortical region</th>
<th>Vehicle (n=8)</th>
<th>Haloperidol (n=7)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACC</td>
<td>1.0 ± 0.1</td>
<td>0.5 ± 0.1</td>
</tr>
<tr>
<td>S1BF</td>
<td>2.0 ± 0.1</td>
<td>1.0 ± 0.1</td>
</tr>
<tr>
<td>PrL/IL</td>
<td>3.0 ± 0.1</td>
<td>1.5 ± 0.1</td>
</tr>
</tbody>
</table>

(All expressed as thousands per mm² ± SEM; n=8; 2 mg/kg/day s.c.)

Figure 2. Operator-independent DBM analysis reveals region specific brain volume changes induced by chronic haloperidol treatment

Figure 3. Validation of DBM results: Manual segmentation and stereology identify a specific reduction in ACC volume following chronic haloperidol treatment

Figure 4. Chronic haloperidol treatment has no significant effect on total neuronal number, but increases neuronal density in the ACC

DISCLOSURE: This poster is supported by project grant funding (G1002198) from the Medical Research Council, UK. Dr Vernon, Miss Chege, Dr Crum, Dr Natesan, Dr Modo, Prof. Cooper, Prof. Williams and Prof Kapur declare no potential conflicts of interest.