Networks of correlated activity in the phMRI response to amphetamine resolved by cluster analysis

A. J. Schwarz[1], A. Gozzi[1], T. Reese[1], and A. Bifone[1]

[1]Neuroimaging, GlaxoSmithKline / Psychiatry CEDD, Verona, Veneto, Italy

Introduction:

Functional MRI methods have been widely used to map the central haemodynamic response following acute pharmacological challenge, but the central effects of drugs can be complex and may include activity at the primary site of action, downstream effects in other brain regions and direct effects on vasculature and neurovascular coupling. Univariate analysis does not discriminate between these effects. However, patterns observed in *correlation* maps referenced to different brain regions [1] hint that the functional response to a drug challenge may recruit different *networks* of brain regions – groups of structures closely coupled to others in the same group but loosely coupled with those in other groups. Here, we present a systematic analysis of the correlation structure underlying the pharmacological MRI (phMRI) response to acute *d*-amphetamine challenge by means of an atlas-defined parcellation of the rat brain and a cluster analysis on the inter-subject response amplitude profiles.

Methods:

Pharmacological MRI (phMRI) data were acquired from male Sprague-Dawley rats under 0.8% halothane anaesthesia at 4.7T using a T_2w RARE sequence and a blood pool contrast agent (Endorem; Guerbet, France) to sensitise the images to relative changes in cerebral blood volume (rCBV) [2,3]. *N*=17 rats received 1mg/kg d-amphetamine i.v. Data were spatially normalised to a stereotaxic rat brain template with associated anatomical atlas [4]. The rCBV time series were then spatially parcellated into time courses from bilateral volumes of interest (VOIs) corresponding to 48 atlas-defined brain structures. The response amplitude in each time course was estimated using general linear model regression [5], resulting in 48 cross-subject response amplitude profiles. These were normalised to zero mean and unit standard deviation and grouped using the *k*-means cluster algorithm [6]. Since this algorithm requires the number of groups to be specified as input, we used the C(g) statistic [6] (essentially the ratio between the inter-cluster and intra-cluster variances) to estimate the optimal number of groups present in the data.

Results:

Fig.1(a) shows the matrix of pair-wise inter-region correlations (thresholded at a false discovery rate of 5%), grouped by anatomical location. The cluster analysis resolved this complex structure into three distinct networks of brain regions, the members of each exhibiting closely coupled responses to acute *d*-amphetamine challenge (Fig. 1(b)):

- Cluster 1 included brain structures involved in the mesolimbic and nigrostriatal dopamine pathways. The midbrain structures from which these projections originate – the VTA and substantia nigra (SN) – are both included, along with ventral structures along the major axes of these pathways (Fig. 2). Structures in this group also had significant connections to the CPu as well as thalamic and pre-frontal cortical brain regions.
- Cluster 2 contained frontal and lateral cortical VOIs along with regions in the pre-frontal cortex (infra-limbic, pre-limbic and cingulate) and sub-cortical structures including the caudate putamen and globus pallidus. These regions shared a widespread correlation pattern reminiscent of the univariate group response.
- Cluster 3 including predominantly midbrain structures including those involved in the periventricular dopamine system. The more dorsal hippocampal VOIs and posterior cortical structures were also included. While these VOIs were predominantly distributed toward the ventral and posterior parts of the brain, they included strong connections to forebrain cortical structures not included in this cluster.

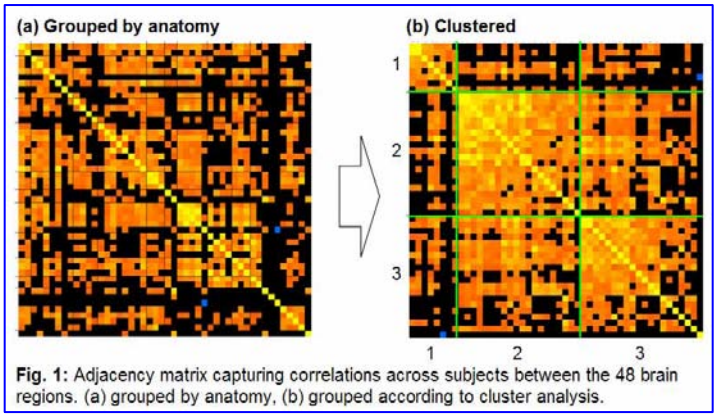

Fig. 1: Adjacency matrix capturing correlations across subjects between the 48 brain regions. (a) grouped by anatomy, (b) grouped according to cluster analysis.

Discussion:

Encouragingly, key brain areas involved in primary dopamine pathways emerged naturally from the data in the form of a group of structures (cluster 1) whose associated correlation patterns is consistent with established anatomical connections including ventral projections from the midbrain to the striatum [7]. This finding confirms the involvement of dopaminergic neuronal systems in the rCBV response to *d*-amphetamine, and shows that this method is able to distinguish meaningful functional relationships in phMRI data. The widespread response in cluster 2 may represent forebrain neuronal activity downstream from that in the primary dopaminergic pathways captured in cluster 1. The components of the response resolved in clusters 2 and 3 may also involve other neurotransmitter systems, such as norepinephrine, which has an extensive innervation of the cortex; either via direct action at the norepinephrine transporter or cross-talk with the dopamine system [8,9]. Alternatively, the presence of responses uncorrelated with those in primary dopamine pathways could be a sign of different mechanisms underlying the rCBV changes. However, we observed no correlation between the amplitude of peripheral blood pressure changes and central rCBV responses in any of the VOIs.

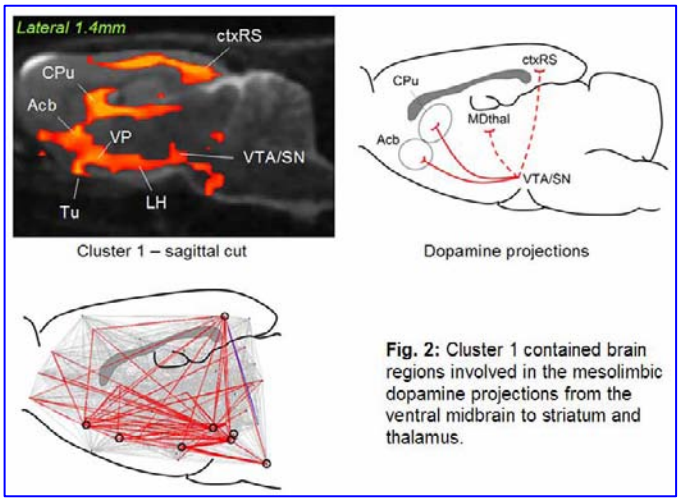

Fig. 2: Cluster 1 contained brain regions involved in the mesolimbic dopamine projections from the ventral midbrain to striatum and thalamus.

Conclusion:

Amphetamine challenge results in a widespread functional connectivity network that was resolved into three distinct sub-networks using a cluster analysis approach. This approach may be useful in disentangling functional relationships in phMRI data and provide important new insights into the central systems underlying pharmacological action.

References:

[1] Schwarz AJ et al. Proc FENS 2006. [2] Schwarz AJ *et al*. (2004) Synapse **54** 1 [3] Gozzi A *et al*. (2006) Neuropsychopharmacology **31** 1690 [4] Schwarz AJ *et al*. (2006) NeuroImage **32** 538. [5] Schwarz AJ *et al*. (2006) J. Neurosci. Meth., to appear. [6] Everitt BJ *et al*. (2001) Cluster Analysis (Arnold, London). [7] Beckstead RM (1976) J. Comp. Neurol. **166**(4) 403 [8] Reith M *et al.* (1997) Psychopharmacology **134**(3) 309 [9] Devoto P *et al*. (2003) Synapse **50**(3) 200.