Brain networks reveal the effects of antipsychotic drugs on schizophrenia patients and controls

Abbreviated title: Brain networks reveal antipsychotic drug effects

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Abstract

The study of brain networks, including derived from functional neuroimaging data, attracts broad interest and represents a rapidly growing interdisciplinary field. Comparing networks of healthy volunteers with those of patients can potentially offer new, quantitative diagnostic methods, and a framework for better understanding brain and mind disorders. We explore resting state fMRI data through network measures. We construct networks representing 15 healthy individuals and 12 schizophrenia patients (males and females), all of whom are administered three drug treatments: (i) a placebo; and two antipsychotic medications (ii) aripiprazole and; (iii) sulpiride. We compare these resting state networks to performance at an "N-back" working memory task. We demonstrate that not only is there a distinctive network architecture in the healthy brain that is disrupted in schizophrenia, but also that both networks respond to antipsychotic medication. We first reproduce the established finding that brain networks of schizophrenia patients exhibit increased efficiency and reduced clustering compared to controls. Our data then reveals that the antipsychotic medications mitigate this effect, shifting the metrics towards those observed in healthy volunteers, with a marked difference in efficacy between the two drugs. Additionally, we find that aripiprazole considerably alters the network statistics of healthy controls. Examining the "N-back" working memory task, we establish that aripiprazole also adversely affects their performance. This suggests that changes to macroscopic brain network architecture result in measurable behavioural differences. This is one of the first studies to directly compare different medications using a whole-brain graph theoretical analysis with accompanying behavioural data. The small sample size is an inherent limitation, and means a degree of caution is warranted in interpreting the findings. Our results lay the groundwork for an objective methodology with which to calculate and compare the efficacy of different treatments of mind and brain disorders.

Introduction

In recent years neuroimaging data and graph theory have allowed for the description of the topological properties of large-scale brain networks (Bullmore and Vértes, 2013; Bullmore and Sporns, 2009; van den Heuvel and Hulshoff Pol, 2010). Disorders of the brain have long been thought to be due to abnormal connectivity patterns, and these networks allow for a quantitative measure of this disruption (Bullmore and Sporns, 2012; Rubinov and Bullmore, 2013). Schizophrenia is a debilitating psychiatric condition with a range of symptoms including auditory and visual hallucinations, delusions, disorganised thinking, and cognitive impairment. Various network-based studies have associated schizophrenia with a subtle randomisation of connections (Alexander-Bloch et al., 2013; Fornito et al, 2012; van den Heuvel et al., 2013; Lynall et al., 2010; Rubinov and Bullmore, 2013; Yu et al., 2011). Antipsychotic medications are employed to treat symptoms with varying degrees of success and side-effect. Sulpiride is a selective dopamine antagonist most commonly used in Europe and Japan for schizophrenia treatment. Aripiprazole is an atypical third generation antipsychotic introduced for the treatment of schizophrenia in the USA in 2002 and Europe in 2004 (Bolstad et al., 2015). It acts as a dopamine receptor partial agonist, whereas typical antipsychotics used to combat the symptoms of schizophrenia are pure dopamine antagonists. Partial agonists have long been of interest (Lieberman, 2004) in order to avoid the extrapyramidal and endocrine side effects caused by typical antipsychotics.

There is a body of evidence demonstrating drug treatments lead to specific localised changes in functional network structure (Cole, 2013; Sarpal et al, 2015), and a growing interest in whole-brain approaches (Carhart-Harris et al, 2017). Yet, few graph theoretical studies to date have been conducted to understand if and how medication alters an individual's whole-brain network (Achard and Bullmore, 2007; Flanagan et al, 2019; Hadley et al., 2016). We hypothesised that an effective medication would act to make the functional brain networks of patients more similar to those of healthy volunteers. We set out to test this in the context of three drug treatments: (i) placebo; (ii) aripiprazole, and (iii) sulpiride. We used resting state fMRI data to analyse the functional connectivity, and a working memory task to assess the cognitive abilities, of 15 healthy volunteers and 12 patients with chronic schizophrenia.

Our results show that schizophrenia patients and healthy controls exhibit different network topologies, in agreement with the existing literature (Alexander-Bloch et al., 2013; Lynall et al., 2010; Pavlović et al, 2019). Further, the antipsychotic drug treatments alter the topology of the brain network in a measurable way, particularly both in healthy individuals. In the brain networks of patients we found evidence that the antipsychotic drugs lead to network topologies that are closer to those of healthy individuals. This correlates with improved cognitive performance. In healthy individuals, treatment with aripiprazole leads to a significantly altered network as well as lower cognitive performance.

Materials and Methods

Experimental Design and Statistical Analysis

Sample

Twelve people with chronic schizophrenia and 15 healthy, nonpsychotic volunteers were recruited for participation in this study (see Supplementary Table 2 for detailed demographics). The patients were diagnosed according to standard operational criteria in the DSM-IV (American Psychiatric Association, 2000) and were clinically stable during their involvement (i.e. exhibiting low symptom ratings and undergoing no change of medication in the preceding four weeks). All were receiving antipsychotic drugs, and four were receiving additional psychotropic medication, but were not treated with their usual medication on the days of scanning to avoid effects on the fMRI data. Healthy volunteers were selected to match the patient group in terms of age, gender, premorbid IQ, years of education, and handedness, and screened for major psychiatric disorders using the Mini International Neuropsychiatric Interview (Sheehan et al., 1998). All subjects provided informed consent in writing and the protocol was approved by the Addenbrooke's NHS Trust Local Research Ethics Committee.

Every subject attended three scanning sessions, each one to two weeks apart, for collection of functional MRI data and completion of working memory tests (see below). At each visit they were administered one of three drug treatments: (i) an oral placebo 180 and 90 mins before scanning; (ii) oral aripiprazole 15mg 180 mins before scanning and oral placebo 90 mins before; (iii) oral placebo 180 mins before scanning and oral sulpiride 400 mg 90 mins before. We used a double dummy design with dosing of aripiprazole 180 mins and sulpiride 90 mins before the start of fMRI scanning. Both patients and experimenters were blind for the drug condition. The study medication was randomised by a colleague, who was not a member of the study team and stored in envelopes for each patient and testing session. There was a sealed envelope with the drug order for each participant that could be opened in case of a serious adverse effect. Both aripiprazole and sulpiride are antipsychotic medications designed to alleviate the symptoms of schizophrenia. At both time points (-180 mins and -90 mins) we co-administered 10mg of domperidone to minimise side effects. Domperidone is a peripheral D2 receptor blocker sometimes used to mitigate nausea in pharmacological functional neuroimaging studies (Nagano-Saito et al., 2009; Ye et al., 2011). It does not cross the blood-brain barrier and will therefore not confound the neuroimaging results. The order of drug administration and the playlists of the working memory paradigm were counterbalanced across one group and repeated for the other.

Working memory tests

At each session, the subjects were required to complete an "N-back" task to assess their verbal working memory (Baddeley, 2003; Koshino, 2005; Owen et al, 2005). The task demanded that subjects maintain a series of visually presented letters in their working memory such that each stimulus could be compared to the letter presented N letters earlier in the series (i.e. N-back) - see Figure 2(a). For example, if the sequence of letters was F-B-A-B, the subject could be expected to indicate on presentation of the last letter in the series that B was presented two letters previously (2-back). Difficulty was manipulated to 4 levels (0-back to 3-back) by varying the number of letters back in the series that the subject had to compare to the current letter. All subjects were provided with written instruction and completed a practice version before undertaking the full task. They completed three playlists matched for difficulty and distraction, which were allocated across sessions such that each subject would complete each playlist once across their three visits (for placebo, aripiprazole, and sulpiride). An individual's performance at this cognitive task was assessed by recording their hit rate, defined as the proportion of times they were able to successfully present a correct answer. In each session, there were 10 correct targets for each N-back level. Data for the performance of patients 11 and 12 administered sulpiride were missing so analyses were carried out without them, leaving $N = 10$ patients and $N = 15$ healthy subjects.

Acquisition and preprocessing of fMRI data

A General Electric (GE) Signa system scanner operating at 1.5 T at the BUPA Lea Hospital (Cambridge, UK)) was used to acquire functional MRI data over 17 min 12s, during which time subjects were asked to lie quietly with their eyes closed. In each session, 516 gradient-echo T2*-weighted echo planar images depicting blood oxygenation level-dependent (BOLD) contrast were generated from 16 noncontiguous near-axial planes: repetition time = 2 s, echo time = 40 ms, flip angle = 70 °, voxel size = 3.05 × 3.05 × 7.00 mm, section skip = 0.7 mm, matrix size = 64 × 64, field of view (FOV) = 240 × 240 × 123 mm. 4 volumes were discarded to allow for T1 equilibration effects, leaving 512 volumes per dataset (Lynall et al., 2010).

Control 2 was missing an anatomical image so was discarded from the study, and patient 11 was missing data for the aripiprazole treatment. Each dataset was analysed for effects of head motion within the scanner (Power et al., 2012; Satterthwaite et al., 2012; Van Dijk et al., 2012), resulting in the further rejection of patient 3 on aripiprazole and sulpiride, control 10 on placebo and sulpiride, control 8 on sulpiride, and patient 5 on placebo, all of which were deemed to have too many motion related artefacts to be reliable (see Supplementary Information). The remaining datasets were corrected for motion through realignment and wavelet despiking (Patel et al, 2014; Suckling et al., 2006). We used a 12 parameter affine transformation to register the data to MNI stereotactic standard space and a 6mm Gaussian kernal for spatial smoothing. Finally, the voxel

timeseries were high- and low-pass filtered with cutoff frequencies of ≈ 0.01 Hz and ≈ 0.08 Hz respectively, as per (Patel et al, 2014; Power et al., 2012). Statistical analyses of subsequent network metrics were performed using those datasets which were available for all drug treatments, equating to 9 patients and 12 controls.

Kolmogorov-Smirnov test

The two-sample *Kolmogorov-Smirnov test* is a non-parametric test to compare two sets of data. The Kolmogorov-Smirnov statistic is a measure of the distance between the empirical distribution functions of the two samples and is calculated under the null hypothesis that both samples are taken from the same continuous probability distribution. The statistic can then be used to assign a p-value to the likelihood that the null hypothesis may be rejected. We used this method to assess the distribution of the global network measures for each of the 6 groups: controls ($\times 3$ - aripiprazole, placebo, sulpiride) and patients ($\times 3$ - aripiprazole, placebo, sulpiride). A p-value of < 0.05 was taken to indicate a significant result.

Analysis of variance (ANOVA)

To examine the effects of the drugs, volunteer type and task difficulty on cognitive performance, we performed a 3 way ANOVA with 2 repeated measures (Cohen, 2007) in various combinations on the hit rates of the subjects. A p-value of < 0.05 was taken to indicate a significant result.

Cohen's d

To quantify effect sizes between groups, we calculated Cohen's d (Cohen, 1988), which is defined as:

$$d = \frac{\bar{x}_1 - \bar{x}_2}{\sigma_{pooled}} \tag{1}$$

where $\bar{x}_1$ and $\bar{x}_2$ are the means of the two samples and $\sigma_{pooled} = \sqrt{\frac{(n_1-1)\sigma_1^2+(n_2-1)\sigma_2^2}{n_1+n_2-2}}$ is the pooled standard deviation for the two samples of size n_1 and n_2.

Games-Howell post-hoc test

The Games-Howell post-hoc test is a modification of the traditional Tukey's Honestly Significant Differences (HSD) test for datasets with unequal variances and/or sample sizes. For two groups with means $\bar{x}_1$ and $\bar{x}_2$ and standard deviations σ_1 and σ_2, the critical q_{crit} can be looked up in the Studentised t statistic q table with a modified degrees of freedom:

$$df' = \frac{(\frac{\sigma_i^2}{n_i} + \frac{\sigma_j^2}{n_j})^2}{\frac{(\frac{\sigma_i^2}{n_i})^2}{n_i - 1} + \frac{(\frac{\sigma_j^2}{n_j})^2}{n_j - 1}} \tag{2}$$

Then we consider the difference between the means, which to be considered significant must satisfy:

$$\bar{x}_1 - \bar{x}_2 \geq q_{crit} \sqrt{\frac{\frac{\sigma_i^2}{n_i} + \frac{\sigma_j^2}{n_j}}{2}} \tag{3}$$

We employ the Games-Howell post-hoc test to examine the drug treatment effects on the patients alone, and use a significant level of 0.05.

Anatomical parcellation and wavelet decomposition

For each individual dataset, voxel time series were averaged within each of the 325 equally sized anatomical regions in a random driven atlas (see (Zalesky et al., 2010) for approach). 28 regions lacked good quality fMRI timeseries for some subjects so were discarded from our analysis, leaving datasets for 297 brain regions for all subjects. The discarded regions are mostly cerebellar, an area known to be highly susceptible to artefacts due to the major arteries which pass in the vicinity (Dagli et al, 1999), and the complete list can be found in the Supplementary Information. The maximal overlap discrete wavelet transform (Percival and Walden, 2000) was used to decompose each individual regional mean fMRI time series into the frequency interval 0.030 - 0.060 Hz (scale 3). This frequency range was selected as it is has been shown that frequencies of ≤ 0.1 Hz exhibit the most prominent salient neuronal fMRI dynamics (Achard et al., 2006).

Topological network construction

Undirected weighted networks were generated for each individual based on correlating scale 3 wavelet coefficients. The resulting correlation coefficients r_{ij} form the weight of the edges connecting regions i and j. A simple thresholding procedure was then applied to eliminate edges with weights smaller than τ; all remaining edges are then given a weight of 1, providing an undirected, unweighted network. The threshold τ can be varied to generate networks with any desired percentage of possible connections. Following the example of (Lynall et al., 2010), which studies a subset of this particular dataset, we choose to focus on $37\% - 50\%$ connectivity. Firstly, this ensures that all graphs are connected, and secondly, it avoids the increasing randomness associated with higher connection densities (Humphries et al., 2006). All results given for the unweighted networks are averages across this range.

Global network measures

Clustering

The *clustering coefficient*, C_i, for a node i can be defined as (Watts and Strogatz, 1998):

$$C_i = \frac{2T(i)}{k_i(k_i - 1)} \tag{4}$$

where $T(i)$ is the number of triangles containing node i, and k_i is the degree of i - see Figure 1(a).

The *average clustering* then provides a global network measure of clustering, and is simply the average of all values of nodal clustering, or:

$$C = \frac{1}{N} \sum_{i \in G} C_i \tag{5}$$

Characteristic path length

If the shortest path lengths, L_{ij}, between all existing node pairs i and j, are identified, then the *characteristic path length* of the network, L, is simply given by the mean of their sum:

$$L = \frac{1}{N(N-1)} \sum_{i \neq j \in G} L_{ij} \tag{6}$$

Efficiency

A measure of the *global efficiency* of a network, E_{Global}, is given by the mean of the sum of the inverse shortest path lengths, L_{ij}, between all existing node pairs i and j (Achard and Bullmore, 2007; Latora and Marchiori, 2001):

$$E_{Global} = \frac{1}{N(N-1)} \sum_{i \neq j \in G} \frac{1}{L_{ij}} \tag{7}$$

where N is the number of nodes in the graph G. Networks for which the average path length is small can thus be said to have high global efficiency (Achard and Bullmore, 2007) (see Figure 1(b)).

This is equivalent to averaging the nodal efficiencies for all nodes in the network.

Assortativity

The *assortativity* of a network is a measure of the preference of its nodes to connect to other nodes of similar degree. Let e_{xy} be the joint probability distribution (or mixing

matrix) of the degrees. Then if $\sum_y e_{xy} = a_x$ and $\sum_x e_{xy} = b_y$ are, respectively, the fraction of edges that start and end at vertices with values x and y and further that $e_{xy} \neq a_x b_y$ (the case of no assortative mixing), the assortativity coefficient can be defined simply by calculating the Pearson correlation coefficient (Newman, 2003):

$$A = \frac{\sum_{xy} xy(e_{xy} - a_x b_y)}{\sigma_a \sigma_b} \tag{8}$$

where σ_a and σ_b are the standard deviations of the distributions a_x and b_y. A has a value in the range $-1 \leq r \leq 1$, where $A = 1$ would correspond to a perfect correlation between x and y, ie perfect assortativity, and similarly $A = -1$ would indicate perfect disassortativity.

Software

Motion diagnostics, preprocessing and parcellation of the functional MRI data were completed using the preprocessing pipeline with temporal despiking from (Kundu et al., 2012; Patel et al, 2014). Metric calculations and network manipulations were carried out using the Python networkx library (Hagberg et al., 2008) and Matlab. We used IBM SPSS Statistics for all ANOVA calculations (IBM Corp., 2012).

Results

The effects of schizophrenia on global efficiency and clustering are mitigated by medication

We first compared the functional brain networks derived from schizophrenia patients and healthy volunteers on placebo treatments, and as expected, (Alexander-Bloch et al., 2013; Lynall et al., 2010) find distinct differences. In agreement with the existing literature, the schizophrenia networks have moderately increased efficiency (median $E_{Global,SZ} = 0.7144$ compared to $E_{Global,HV} = 0.7132$, $p = 0.04$, $d = 0.38$) and a large reduction in clustering (median $C_{SZ} = 0.6765$ compared to $C_{HV} = 0.7079$, $p = 0.01$, $d = 1.06$) - see Figure 1. With clustering and efficiency values intermediate between those of random graphs and lattices, both the healthy and patient networks exhibit small-world properties (Liu, 2008).

We next employed an ANOVA (two-way, one repeated measure) to examine any differences in network measures between groups, and the factors underlying them. This confirmed group differences due to subject type on both efficiency and clustering ($p = 0.010$ and $p = 0.002$ respectively), and also indicated group differences due to drug effect ($p = 0.041$ and $p = 0.05$ respectively) - see Tables 1 and 2, and Supplementary Table 2. Our hypothesis was that the antipsychotic medications would aim to make the brain connectivities of patients more similar to those of healthy individuals. In the light of the observed differences between the control and patient placebo groups, this hypothesis translates to an expectation that aripiprazole and sulpiride will reduce efficiency and increase clustering. We do indeed find this for the healthy volunteers, but only small such changes for the patients (Figure 1). Moreover, after each antipsychotic drug treatment the brain networks of people with schizophrenia had global efficiencies which were no longer statistically different from the healthy brain networks (median $E_{Global} = 0.7137$, $p = 0.241$, $d = 0.16$ for aripiprazole and $E_{Global} = 0.7135$, $p = 0.081$, $d = 0.37$ for sulpiride). Both antipsychotic drugs also led to average clustering coefficients that were much closer to those of healthy brain networks (median $C = 0.6867$, $p = 0.032$, $d = 0.83$ for aripiprazole, $C = 0.6952$, $p = 0.081$, $d = 0.60$ for sulpiride). Post-hoc tests revealed that differences in efficiency and clustering across drug treatments in patients alone are not significant (see Supplementary Table 4).

Aripiprazole significantly changes healthy brain networks

We found that aripiprazole has a dramatic effect on healthy individuals, with a large variation across individuals. We observe significantly reduced global efficiencies (median $E_{Global} = 0.7080$, $\sigma = 0.009$, $p = 0.006$, $d = 1.08$), and an even greater increase in clustering (median $C = 0.7490$, $p<0.001$, $d = 1.63$). Sulpiride increased clustering in healthy networks (median $C = 0.7294$, $p = 0.028$, $d = 0.74$), but had no significant effect on global efficiency (median $E_{Global} = 0.7094$, $p = 0.321$, $d = 0.43$). Almost all metrics ex-

amined are greatly altered in the healthy volunteers administered aripiprazole, indicating considerable restructuring of functional connectivity: see Figure 1 and Supplementary Table 2. These results are consistent with observations in the brain networks of people with schizophrenia: the drug treatments reduce efficiency and increase clustering.

Cognitive performance of healthy individuals is impaired after taking aripiprazole

All subjects score consistently highly on the very easy 0-back task, but their performance deteriorates considerably with increasing difficulty of the task, with subjects experiencing profound difficulty with the 3-back version (see Figure 2, Table 2, and Supplementary Table 3). In the healthy cohort, aripiprazole has a detrimental effect on performance, whereas the impact of sulpiride is negligible - see Figure 2(b). In the challenging 2-back version of the task, the disparity is most clear - aripiprazole has a negative impact on cognitive performance, giving rise to an average hit rate of 0.65 ± 0.21 (compared to 0.83 ± 0.19 on placebo). Sulpiride, however, has no noticeable impact, with subjects achieving an average hit rate of 0.83 ± 0.18.

An ANOVA (three-way, two repeated measures) revealed that, naturally, the predominant factor in determining success at the working memory tests was the difficulty of the task ($p<0.0001$) - see Table 3. However, it also demonstrated that the drug treatments have a significant effect ($p = 0.007$). We then separated out the drug treatments into placebo-aripiprazole and placebo-sulpiride groups and repeated the ANOVA on controls and patients separately. This shows that the effect is only observed in the healthy volunteers, and that aripiprazole is the medication responsible ($p = 0.015$, $d = 0.37$for the placebo-aripiprazole groups and $p = 0.769$, $d = 0.04$for the placebo-sulpiride groups). Thus, amongst healthy volunteers, we find that aripiprazole results in poorer performance at the N-back working memory task as compared to placebo, and that sulpiride has no noticeable effect.

Cognitive tests show worse performance of patients but do not capture drug effects

Patients scored worse than controls in the N-back working memory task, but not significantly so, with an average hit rate of 0.73 ± 0.18 for patients and 0.83 ± 0.19 for controls for the placebo 2-back task. Aripiprazole and sulpiride do not change the performance of patients much (average hit rates of 0.71 ± 0.21 and 0.79 ± 0.28 respectively for the 2-back task) with an ANOVA showing no significant drug effect ($p = 0.217$) - see 2(c).

Discussion

Network topology, illness, and medication

It has been known for some time that the whole-brain functional brain network organisation of people with schizophrenia differs from that of healthy volunteers (Alexander-Bloch et al., 2013; Lynall et al., 2010). However, very few graph-theoretic studies have been conducted into the effect of antipsychotic medication, or indeed any drug for any brain disorder, on this network organisation. The ones that have been conducted found measurable drug effects (Achard and Bullmore, 2007; Flanagan et al, 2019; Schmidt et el., 2013; Yang et al., 2014) (including, converse to treatment, inducing psychosis (Lahti et al., 2001)). We hypothesised that a drug designed to treat schizophrenia would modify the brain connectivities of patients, making them more similar to those of healthy individuals. The results for global efficiency and clustering in Figure 1 clearly demonstrate this principle - sulpiride and aripiprazole act to reduce efficiency in both controls and patients. While the differences among the patient group do not prove to be significant, they do leave the patients with network efficiencies and clustering comparable to those of the unmedicated controls, and the controls have lower efficiency and higher clustering than before. In addition we also observed a strong effect of a single dose of aripiprazole on medication naïve healthy controls. This finding is consistent with our result in patients, as the drug seemingly tries to 'correct' for schizophrenia network characteristics - in the absence of schizophrenia - by altering the network metrics to decrease efficiency and increase clustering..

Network topology and cognitive ability

Our results suggest that there is an optimal configuration for a brain network in terms of maximising cognitive ability: performance worsens given any change (increase *or* decrease) in the examined metrics from a healthy baseline, the control placebo group. We saw that as well as having a characteristically different brain network structure, schizophrenia patients perform less well at tests of cognitive ability than their healthy counterparts, as has been previously demonstrated (Bullmore and Sporns, 2012), although these effects are not significant. Further, we showed that the group who performed most differently on medication (healthy volunteers having been administered aripiprazole) was also the group who had the most changes to the topology of their brain networks. This supports the notion that one's cognitive ability is intrinsically linked to the structure of the brain's functional network (Basset et al., 2009). For example, to integrate and process information quickly, a network requires some level of efficiency. The control group on aripiprazole had diminished efficiency and performed significantly worse at the N-back tasks than when on placebo. No such impaired performance is seen for sulpiride, for which the reduction in efficiency was negligible. On the other hand, the schizophrenia brain networks

appear to have *too* high an efficiency, perhaps leading to disordered or overwhelming information integration, and they too perform worse. The drugs do non-significantly decrease efficiency in patients and their performance is slightly improved at the 1- and 2-back tasks. A previous study on MEG derived networks using the N-back working memory paradigm (Kitzbichler et al., 2011) demonstrated a shift towards a more random network configuration (with a decrease in modularity and clustering, and an increase in global efficiency) as the cognitive demands of the task increased. The authors also note significant differences from purely random networks and argue that global sychronisation is important in higher cognition, which is reflected in the network architecture.

Neurochemical differences in schizophrenia

Schizophrenia implicates D2 receptors (among others) (Howes et al., 2009; Morgan et al., 2019) and a succession of studies have demonstrated increased presynaptic dopamine synthesis in psychosis and at risk patients (Howes et al., 2009, 2011). Dopamine antagonists such as aripiprazole and sulpiride are therefore used to treat schizophrenia. PET studies of schizophrenia patients have found that greater dopamine receptor occupancy by aripiprazole was associated with better working-memory performance in terms of error rate and reaction time (Shin et al., 2018). Conversely, in healthy volunteers greater striatal D_2 receptor occupancy by aripiprazole was related to greater decrease in frontal metabolism, and greater reduction in frontal metabolism was associated with impaired performance at a working memory task (Kim et al., 2013). Thus striatal dopaminergic function could contribute to the working-memory impairments observed in schizophrenia, and antipsychotic drugs could mitigate this by reducing excess striatal dopaminergic neurotransmission. With no excess dopamine synthesis in healthy volunteers, a reduction induced by dopamine antagonists leads to worsened performance.

Consistent with the literature, our results indicated that aripiprazole significantly worsened the performance of healthy subjects at the N-back working memory task, but did not hinder the patients. Sulpiride had no significant detected effects. The main pharmacological difference between the two antipsychotics is that aripiprazole is a partial D_2 antagonist and sulpiride is the most selective D_2 antagonist. Given this we might expect that sulpiride have a larger detrimental effect on the healthy controls' performance, but it is possible that due to the high dose of aripiprazole it had stronger $D2_2$ antagonistic effects than the single dose of sulpiride. It is also surprising that we observed such small differences between the healthy and patient groups at the working memory tasks. However, we recruited relatively stable and high functioning patients, and all participants were trained outside the scanner so that their performance was relatively stable. We aimed to match patients and controls for task performance so that drug effects were not confounded by group effects at the baseline (the placebo treatment).

Limitations

The greatest limitation of this study is the small sample size: $N = 15$ for healthy controls and $N = 12$ for patients with schizophrenia, which is further reduced to $N = 12$ and $N = 9$ respectively for the functional brain networks, and $N = 15$ and $N = 10$ respectively for the working memory tasks. The fMRI timeseries were collected with standard parameters at the time of study, which given the age of the datasets, inevitably have some drawbacks compared to modern state of the art datasets. Most notably, this includes a greater sparsity of imaging, compounded by the loss of 28 regions due to head motion artefacts. However, we do note that the acquisition time was long (17 mins 12s), which will have benefited the accuracy of the regional correlations and therefore also the networks we derived from them (Achard et al., 2008). There is a considerable literature comprising studies based on this type of data (Liu, 2008; Lynall et al., 2010), and importantly, key network-based results have been replicated in more modern studies (Hadley et al., 2016; Nelson et al., 2017).

Finally, there are likely confounding effects from other drugs. All patients were prescribed oral antipsychotic medication and asked not to take their medication on the days of the fMRI study (see Figure 1-2 for the details of their usual medications). While this mitigates acute pharmacological effects, patients had been treated with these other antipsychotics for several years and their long-term effects cannot be accounted for. This limitation affects the analysis and interpretation of group effects (differences between patient and controls), but not for the comparison of drug conditions within each group (of patients and controls). An earlier study utilising a subset of this data (Lynall et al., 2010) found no significant correlation with antipsychotic dosage (in chlorpromazine equivalents) and any of the connectivity or network metrics they examined.

Brain networks as a means to assess medication

This quite unique dataset allowed for an investigation into the effects of antipsychotic drugs on both the large-scale functional brain networks and the cognitive performance of people diagnosed with chronic schizophrenia and healthy volunteers. Despite its limitations, clear drug effects were observed on the network topology and performance at the N-back working memory task. We find a reduction in the difference between specific healthy and patient network metrics, and that aripiprazole impairs cognitive ability and radically rewires the brain networks of healthy volunteers. It would be highly beneficial for future studies to use state of the art functional MRI data to further investigate the links between disrupted networks in people with brain disorders, how medication influences these, and an "ideal" network topology for the brain (which should be identified by association with an optimal behavioural parameter, such as the best cognitive performance). Such advanced studies have the potential for not just diagnosis of the original brain disorder, but also for the quantification of the effectiveness of a drug in treating the illness.

This would then allow for a systematic comparison between alternative treatments.

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Legends

Figure 1 : Global efficiency and average clustering values. The box plots display distributions of (a) average clustering and (b) global efficiency for the networks of each group and drug, for individuals with networks for every drug treatment. The extreme ends of the whiskers correspond to the maxima and minima and the white line in the box corresponds to the median. Controls ($N = 12$) are grouped on the left and patients ($N = 9$) on the right. Placebo is shown in blue, aripiprazole in pink, and sulpiride in gold. P-values refer to likelihood the distributions match that of the control placebo group (2-sample K-S test). Outliers are defined as being more than 1.5× the interquartile range away from the median; note this is purely visual and no values are excluded from statistical analyses. Schizophrenia is associated with lower clustering and higher efficiency, seen by comparing the control placebo plot to the patient placebo plot. The antipsychotic medications increase clustering and decrease efficiency, therefore moving patients closer to controls and affecting the control networks. All values and the demographics of all participants can be found in the Supplementary Data Sheets. The schematics illustrate the concepts of (a) clustering and (b) efficiency. (a) Clustering measures the number of triangles which exist around around a node (green solid lines), as a proportion of those that could (also green dashed lines). (b) Efficiency averages the inverse shortest paths (green lines) between all node pairs; many short paths equates to higher efficiencies.

Figure 2 : N-back working memory task. Panel (a) illustrates the nature of the task. Subjects are shown a sequence of letters, and asked to indicate if they are presented with a letter which matches the letter presented "N-back". In the example shown, the subjects are expected to note for the 2-back task that B was presented two letters previously (and for the 0-back task that they are observing the current letter). There is nothing to note for the 1-back and 3-back tasks. Naturally, 0-back is the easiest version of the task and 3-back the hardest. For each drug treatment, hit rates (or fraction of correct responses out of a total of 10 prompts) from (b) healthy individuals ($N = 15$) and (c) schizophrenia patients ($N = 10$) averaged across each level of difficulty are presented as box plots. Results for placebo are shown in blue, aripiprazole in pink, and sulpiride in gold. The extreme ends of the whiskers correspond to the maxima and minima and the white line in the box corresponds to the median.Values below the boxes represent the median values, and for the drug treatment groups, the difference with the median of the placebo group is provided in brackets. Aripiprazole is associated with a reduced number of correct answers as compared to placebo, most strikingly so for controls completing the 2-back task. All values can be found in the Supplementary Data Sheets.

Table 1: Summary statistics for a 2 way ANOVA with 1 repeated mea-

sure on the network global clustering values of patients and healthy controls treated with placebo, aripiprazole, and sulpiride. Individuals for which networks were available for all drug treatments were used, equating to $N = 12$ for healthy controls and $N = 9$ for patients. There is a significant difference between the network clustering of the HV and SZ groups ($p = 0.002$), a significant drug effect ($p = 0.005$) and an additional drug-group type interaction term ($p = 0.005$) - all highlighted in red. This interaction term stems from the effect of aripiprazole - it greatly increases the clustering of control networks whilst causing only a small and variable increase in the schizophrenia networks. The placebo and sulpiride treatments have a more consistent effect on the two groups.

Table 2: Summary statistics for a 2 way ANOVA with 1 repeated measure on the network global efficiency values of patients and healthy controls treated with placebo, aripiprazole, and sulpiride. Individuals for which networks were available for all drug treatments were used, equating to $N = 12$ for healthy controls and $N = 9$ for patients. There is a significant difference between the network efficiency of the HV and SZ groups ($p = 0.010$) and a significant drug effect ($p = 0.041$) - highlighted in red.

Table 3: Summary statistics for a 3 way ANOVA with 2 repeated measures on the hit rates during a working memory task with 4 levels of difficulty of patients and healthy controls treated with placebo, aripiprazole, and sulpiride. Individuals for which data were available for all drug treatments were used, equating to $N = 15$ for healthy controls and $N = 10$ for patients. We see a significant effect of cognitive difficulty ($p < 0.001$) and a significant drug effect ($p = 0.007$) - highlighted in red.

Tables and Figures

Table 1. Summary statistics for a 2 way ANOVA with 1 repeated measure on the network global efficiency values of patients and healthy controls treated with placebo, aripiprazole, and sulpiride.

Source	SS	df	MS	F	p
Between groups					
Subject type	< 0.001	1	< 0.001	8.093	0.010
Error	0.001	19	< 0.001		
Within groups					
Drug	< 0.001	2	< 0.001	3.480	0.041
Drug*Subject type	< 0.001	2	0.0001	1.005	0.376
Error	0.0010	38	< 0.001		

Table 2. Summary statistics for a 2 way ANOVA with 1 repeated measure on the network global clustering values of patients and healthy controls treated with placebo, aripiprazole, and sulpiride.

Source	SS	df	MS	F	p
Between groups					
Subject type	0.026	1	0.026	12.208	0.002
Error	0.040	19	0.002		
Within groups					
Drug	0.004	2	0.002	6.023	0.005
Drug*Subject type	0.004	2	0.002	5.997	0.005
Error	0.012	38	< 0.001		

Figure 1. Average clustering and global efficiency values.

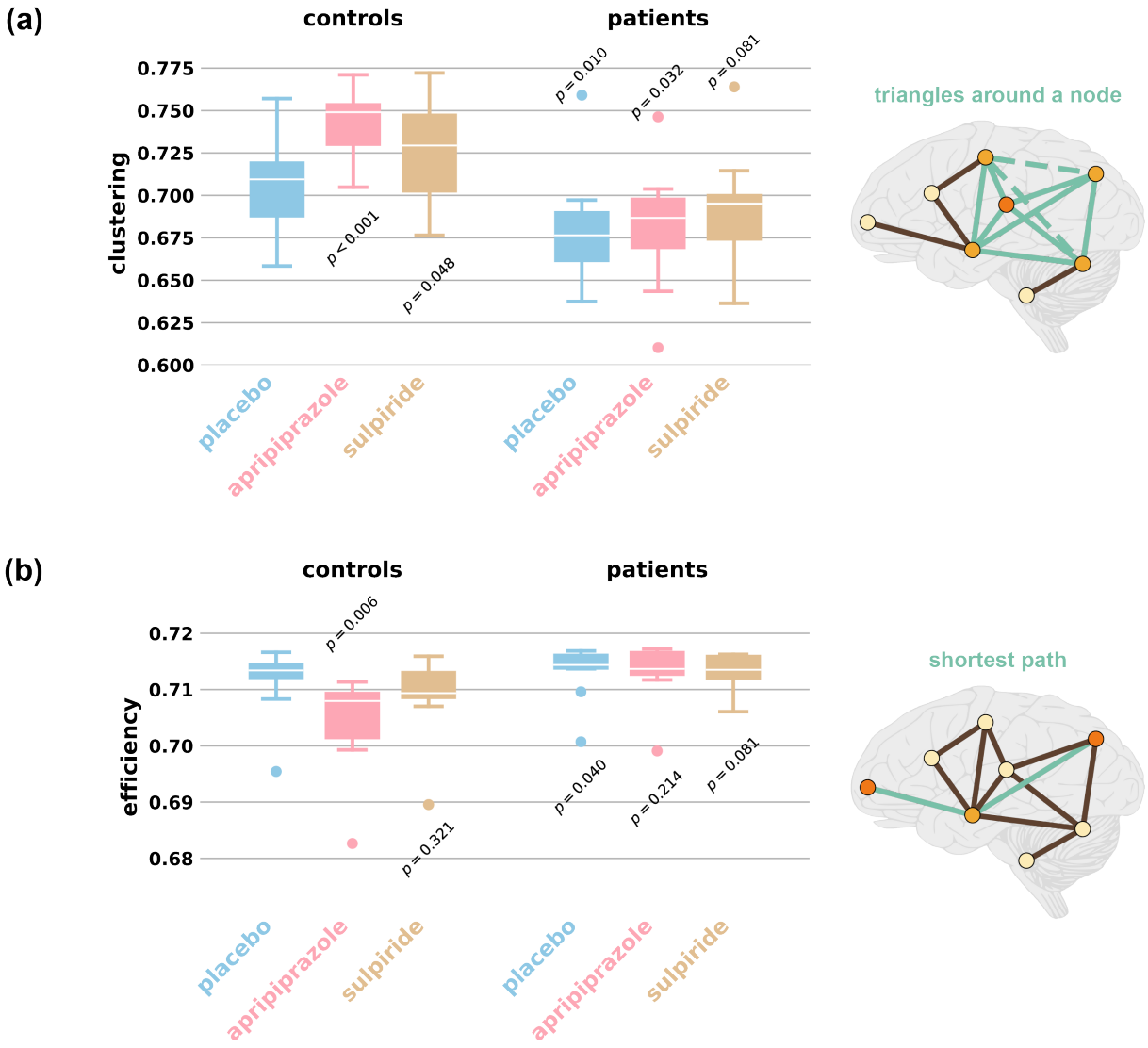

Figure 2. N-back working memory task.

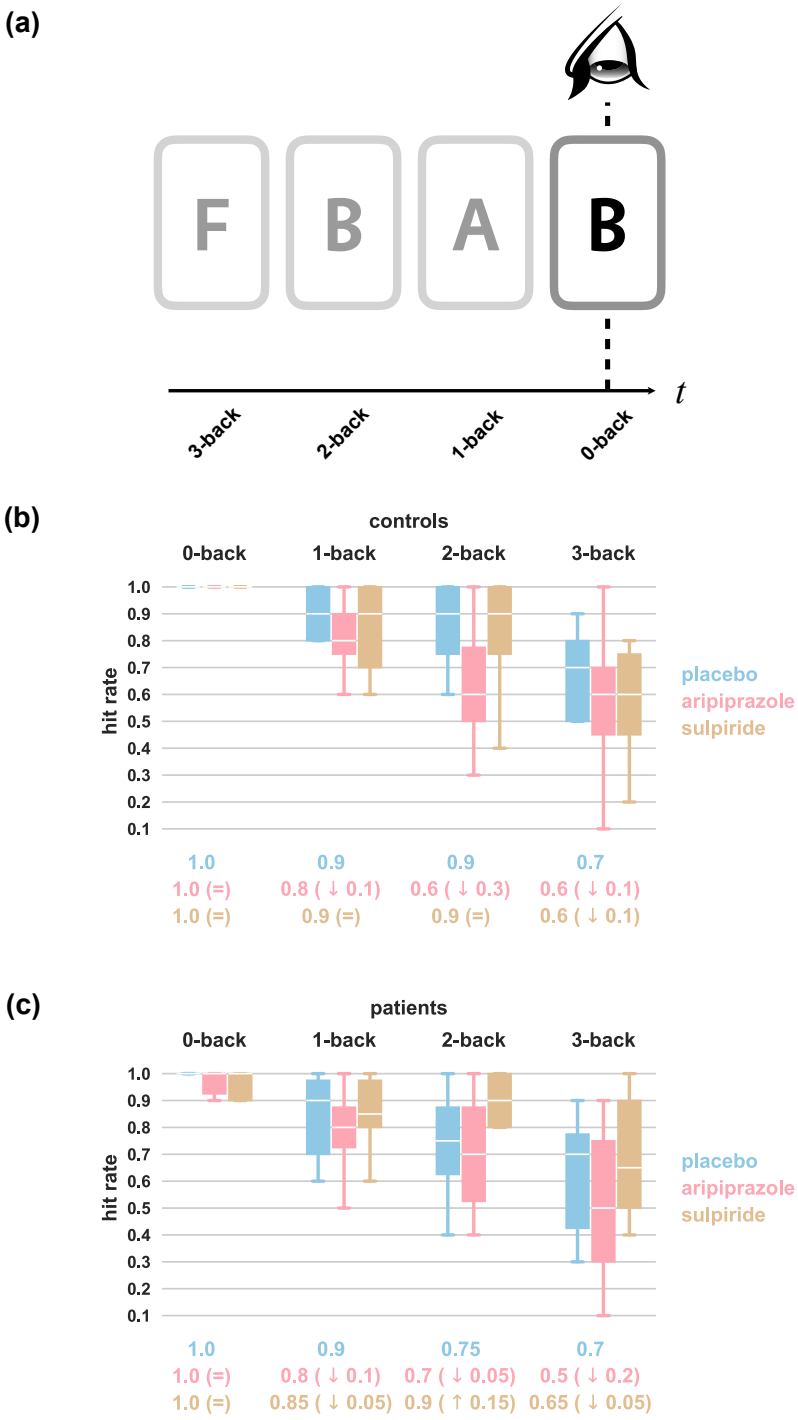

Table 3. Summary statistics for a 3 way ANOVA with 2 repeated measures on the hit rates during a working memory task with 4 levels of difficulty of patients and healthy controls treated with placebo, aripiprazole, and sulpiride.

Source	SS	df	MS	F	p
Between groups					
Subject type	0.007	1	0.007	0.044	0.836
Error	3.762	23	0.164		
Within groups					
Drug	0.386	2	0.193	5.489	0.007
Drug*Subject type	0.011	2	0.006	0.160	0.853
Error	1.618	46	0.035		
Cog. difficulty	4.828	3	1.609	42.779	<0.0001
Cog. difficulty*Subject type	0.045	3	0.015	0.403	0.751
Error	2.596	69	0.038		
Drug*Cog. difficulty	0.120	6	0.020	1.160	0.331
Subject type*Drug*Cog. difficulty	0.162	6	0.027	1.160	0.331
Error	2.384	138	0.017		