Investigation of abnormalities of glutamatergic neurotransmission and cortical function in schizophrenia using MRS at ultrahigh field (7T) and Magnetoencephalography (MEG)

Schizophrenia Research Fund & Mental Health Research UK Joint Award

John Grace QC PhD Scholarship 2013, University of Nottingham, Institute of Mental Health

Supervisors: Professor Peter Liddle (Psychiatry) and Professor Peter G Morris I(Physics)

Research Student: Jyothika Kumar

Hi! My name is Jyothika, and I am very glad to be starting my PhD this year at the Division of Psychiatry, University of Nottingham, all thanks to MHRUK and the Schizophrenia Research Fund for awarding me the John Grace QC PhD Scholarship. I am supervised by Professor Peter Liddle, Professor Peter Morris and Dr. Lena Palaniyappan, and my research project involves using multi-model brain imaging techniques to investigate the relationship between glutamatergic abnormalities and cortical activity in Schizophrenia. I am certain that this PhD is going to be an exciting challenge, and I hope to gain a wide range of research skills in the upcoming 4 years. I am very much looking forward to working with a fantastic team of researchers here and to contribute to the outstanding translational mental health research undertaken at the University of Nottingham. Neuroimaging techniques have helped us understand a lot about brain structure and function, and it would be great to be able to translate all this information into clinical practice and to help improve outcomes in people with mental health illnesses. I do hope my research can play a small but important role in this process.

Start date: September 2013

Summary

Existing treatments for schizophrenia are moderately effective in treating acute symptoms such as delusions and hallucinations, but these treatments are largely ineffective in improving disabling difficulties with memory and concentration. These cognitive functions depend on the efficient transmission of information in networks of brain cells. The major chemical messenger that transmits messages from one cell to another in these networks is glutamate. The function of glutamate is abnormal in schizophrenia. During the past decade, one of the most promising lines of research into new treatments for schizophrenia has been investigation of treatments acting on glutamate. However this research has been hampered

because of a serious lack of detailed knowledge about the nature of the abnormality of glutamate. The problem is made difficult because brain imaging techniques using conventional MRI scanners cannot distinguish clearly between glutamate and a closely related brain molecule glutamine. Distinguishing glutamate from glutamine non-invasively in the living human brain requires an ultrahigh field (7T) MRI device. Such devices are only available in a few highly specialised centres world-wide and have not hitherto been used to measure glutamate in schizophrenia. We currently have funding for a comprehensive study of the brain in schizophrenia using the 7T MRI device and other imaging resources in Nottingham. The student will join the team engaged in this study and take responsibility for addressing the specific question regarding the nature of the imbalance of glutamate and its effect on the function of brain networks.

Article | OPEN | Published: 22 June 2018

The full text of this article, can be seen at: **https://www.nature.com/articles/s41380-018-0104-7**

Glutathione and glutamate in schizophrenia: a 7T MRS study

Abstract

In schizophrenia, abnormal neural metabolite concentrations may arise from cortical damage following neuroinflammatory processes implicated in acute episodes. Inflammation is associated with increased glutamate, whereas the antioxidant glutathione may protect against inflammation-induced oxidative stress. We hypothesized that patients with stable schizophrenia would exhibit a reduction in glutathione, glutamate, and/or glutamine in the cerebral cortex, consistent with a post-inflammatory response, and that this reduction would be most marked in patients with “residual schizophrenia”, in whom an early stage with positive psychotic symptoms has progressed to a late stage characterized by long-term negative symptoms and impairments. We recruited 28 patients with stable schizophrenia and 45 healthy participants matched for age, gender, and parental socio-economic status. We measured glutathione, glutamate and glutamine concentrations in the anterior cingulate cortex (ACC), left insula, and visual cortex using 7T proton magnetic resonance spectroscopy (MRS). Glutathione and glutamate were significantly correlated in all three voxels. Glutamine concentrations across the three voxels were significantly correlated with each other. Principal components analysis (PCA) produced three clear components: an ACC glutathione–glutamate component; an insula-visual glutathione–glutamate component; and a glutamine component. Patients with stable schizophrenia had significantly lower scores on the ACC glutathione–glutamate component, an effect almost entirely leveraged by the sub-group of patients with residual schizophrenia. All three metabolite concentration values in the ACC were significantly reduced in this group. These findings are consistent with the hypothesis that excitotoxicity during the acute phase of illness leads to reduced glutathione and glutamate in the residual phase of the illness.

I am pleased to tell you that we have been awarded the 2019 Margaret Temple award for research into schizophrenia by the BMA Foundation for Medical Research. It's very exciting, my first ever research grant and first project as a Principal Investigator! We will be recruiting and scanning around 100 patients with schizophrenia with varying clinical symptoms in order to identify which patients will be best suited to a specific type of intervention. More details here: http://www.bmafoundationmr.org.uk/grant-winners-2019

It's not a huge amount (£63,950) but its so great that external reviewers and a panel thought that our work and ideas were important and worth pursuing. Also, they only award one of these each year so its amazing to receive it! The BMA will be hosting an awards ceremony in London in November where we (me and co-applicant) will receive the award certificates along with the winners in other categories.. :)

I wanted to share this success with you because the project directly builds on the work that I did during my PhD (the Molecular Psychiatry paper). I'm so grateful for the funding and support I received from MHRUK for my PhD. None of this would have been possible without that! :)

Thanks so much once again!

Jyothika's Final Report

I submitted my thesis titled 'Multimodal Neuroimaging of the Salience Network in Schizophrenia' in September 2017 and graduated with a PhD in Psychiatry from the University of Nottingham in July 2018.

As the title suggests, the main aim of this work was to examine the neurobiological basis of Salience Network dysfunction in patients with schizophrenia using different types of non-invasive brain imaging techniques. One of the main questions I have spent time investigating is whether certain neurochemicals involved in long-range communication (i.e. glutamate) and protection against neuroinflammation (glutathione) are affected in schizophrenia and whether are related to the level of symptoms or dysfunction experienced by patients. This work was published (finally!) this year in a really good journal (Molecular Psychiatry), a really nice graduation present! I am still working on other papers from the work I did during my PhD, and I hope these will be published over the next year. These relate to investigating abnormalities in the function and connectivity of the Salience Network.

Overall, this PhD has been a fantastic learning experience for me. I have realized that I truly enjoy research, and I am very keen on continuing my work in the field of mental health and neuroimaging research. I am currently working as a Research Fellow in the same

department – Centre for Translational Neuroimaging, Division of Psychiatry, University of Nottingham. I am working on a few projects, some are directly related to the work I did during my PhD, but others relate to understanding the effects of cognitive and other types of interventions on the brain, which is quite exciting. This has given me the opportunity to develop more diverse sets of skills and work with different populations. We have recently applied for some funding to continue to do this type of research and I am going to start working on fellowship applications soon. Very much looking forward to the next stages of my research career!

2017 Report

This final year of my PhD has been very exciting. It is almost unbelievable that it has been 4 years since I started working on this project! This past year, I have mainly spent time finalising all my results and doing some last analyses in order to wrap up my main research chapters.

One of the main questions I have spent time investigating is whether certain neurochemicals involved in long-range communication (i.e. glutamate) and protection against neuroinflammation (glutathione) are affected in schizophrenia and whether these would be related to the level of symptoms or dysfunction experienced by patients at the time of scanning. Our results showed that levels of these neurochemicals (measured using a noninvasive imaging technique called MR spectroscopy) were altered in patients who were suffering from residual schizophrenia – a chronic phase in the development of the illness where patients continue to suffer from negative symptoms and socio-occupational dysfunction, although positive symptoms are no longer present. We also found an interesting association between glutathione and glutamate, showing that there is a mechanistic link between these two systems. This raises interesting questions about how these two systems interact with each other in healthy individuals and how they are affected in schizophrenia. We had submitted this manuscript last year for publication but after a couple of rejections, we are now revising it after review for a top tier journal – Molecular Psychiatry. This work is going to make up the first research chapter in my thesis. My second research chapter will look at the functioning of the salience network (a set of brain regions involved in deciding whether any stimulus or event is relevant or not) in schizophrenia. Our group has developed this novel and interesting task called the relevance modulation task to engage the salience network and in this chapter, I will present the results from this task in two modalities – Magnetoencephalography (MEG) and functional MRI (fMRI). We have some interesting results which tie in with previous theories of inefficient brain recruitment in schizophrenia. In my last research chapter, I will look how regions within the salience network interact with each other using causal connectivity analysis in fMRI and then relate these findings to abnormalities in glutamate and

glutathione (mentioned above), which I have reported in my first chapter. So, this chapter will tie all my three research chapters together.

My hope is to complete the first draft of the thesis over the next 6 to 8 weeks and I will be submitting it in September. I have worked on many more things over the past 4 years but for the thesis, I have decided to restrict myself to three main research chapters in which I aimed to answer three key research questions. I think the results make important contributions to brain imaging research in schizophrenia and I am very proud of the work that I have done. Apart from data analysis and thesis writing, I have not been involved in very much over the past year. However, I attended an International conference this year in April – the International Society for Magnetic Resonance in Medicine (ISMRM) which was held in Honolulu, Hawaii. It was a great learning experience and I came back with a lot of exciting ideas for future work.

Overall, this PhD has been a fantastic experience and I would really like to thank MHRUK for providing the funding that made all of this possible! There have been many ups and downs, which is as expected, but I am really glad that I decided to do this PhD and I think I picked the best place to do it. My supervisors have been very supportive and I couldn't have asked for better mentors. Over the past few years, I have developed a strong interest in continuing to work in the field of mental health research and I will be applying for external fellowships next year. In the coming years, I hope that I can play a role, even if it's only a small one, in improving our understanding of how the brain (and the mind) works in health and disease.

2016 Report

My PhD project is a part of two larger research studies conducted by an inter-disciplinary team of researchers here at Nottingham. The first study is called Multimodal Imaging Study in Psychosis (MISP) and was designed to investigate differences in the structure and function of the brain between people with a diagnosis of schizophrenia or bipolar disorder who have also experienced a psychotic episode, and healthy volunteers.

In this study, one of the main questions we were interested in investigating was whether certain neurochemicals involved in long-range communication (i.e. glutamate) and protection against neuroinflammation (glutathione) are affected in schizophrenia and whether these would be related to the level of symptoms or dysfunction experienced by patients at the time of scanning. I have been working on this over the past year and we found that levels of these neurochemicals (measured using a non-invasive imaging technique called MR spectroscopy) were altered in patients who were suffering from residual schizophrenia – a chronic phase in the development of the illness where patients continue to suffer from negative symptoms and socio-occupational dysfunction, although positive symptoms are no longer present. We also found an interesting association between glutathione and glutamate, showing that there is a mechanistic link between these two systems. This raises interesting questions about how these two systems interact

with each other in healthy individuals and how they are affected in schizophrenia. I gave a talk about these results at the International Society of Magnetic Resonance in Medicine (ISMRM) annual conference held in Singapore this year in May and the audience was very interested in these findings. We have now submitted this manuscript for publication and are awaiting the results of the peer review process.

I have also been working on analysing other imaging data from the MISP study. For instance, I am using structural MRI images to analyse whether there are abnormalities in brain structure in schizophrenia and how these might relate to the neurochemical abnormalities that we have discovered in this sample. I have also been working on functional MRI data, looking at how important brain networks function in patients with schizophrenia compared to healthy controls. Preliminary results look interesting but these analyses need further processing in order to be able to fully understand the implications of findings resulting from this work. This is all very exciting as I think these results will tie in nicely with the findings we already have from the MR spectroscopy and will allow me to present a nice, coherent thesis.

Over the last year, we also completed data collection for another research study which aims to develop a protocol to assess the effects of treatment on abnormal brain networks in mental disorders. Healthy volunteers were tested on two days and we will compare patterns of brain activity during task performance and at rest, and neurochemicals in key brain network nodes, following the administration of galatamine with similar observations following administration of the matched placebo. I am working on analysing the data from this study as well now. Potential questions include: how does galantamine affect key brain networks during task and at rest? Does galantamine administration lead to a change in the concentration of neurochemicals?

I aim to spend the next few months finishing these analyses and hopefully getting another manuscript or two ready for submission. I will move into my thesis pending year in October and by the end of this year I will start focusing on wrapping up my thesis and getting it ready for submission. Busy but exciting times ahead!

2015 Report

“My PhD project is a part of two larger research studies conducted by an inter-disciplinary team of researchers here at Nottingham. The first study is called Multi-modal Imaging Study in Psychosis (MISP) and was designed to investigate differences in the structure and function of the brain between people with a diagnosis of schizophrenia or bipolar disorder who have also experienced a psychotic episode and healthy volunteers. When I started my PhD in October 2013, the data for this study were already being collected. So I spent the first couple of months receiving training in the various aspects of data collection, such as how to operate the scanners, participant recruitment, doing clinical interviews etc. Most of the last year has been spent collecting data for this study and also doing some

preliminary analyses. Past research has shown that there are widespread abnormalities in the way various brain regions and networks interact with each other in people with schizophrenia. Yet, we do not know how these abnormalities are related to the symptoms or other factors associated with the illness. These are some of the research questions that I am going to address in my thesis. Which brain networks are abnormal in schizophrenia? How is this related to other brain features? How is this related to symptoms and the clinical picture? Do brain networks behave differently in people with schizophrenia and bipolar disorder? Some symptoms are often treatment-resistant and cause a significant level of dysfunction. Psychiatric medicines work by targeting neurochemicals in the brain, as abnormally high or low levels of these chemicals are thought to be one of the causes of the illness. In this study, we are also interested in investigating whether certain neurochemicals involved in long-range communication such as glutamate and GABA are affected in schizophrenia and bipolar disorder with psychosis and whether any such abnormalities could relate to or predict functioning of important brain networks. As this is a multi-modal imaging study, we are gathering data about the structure and function of the brain using different neuro-imaging techniques such as structural and functional Magnetic Resonance Imaging, Magnetoencephaography and Magnetic Resonance spectroscopy, each of which give us different types of information which we can look at independently and also in conjunction with the other methods. We have some exciting preliminary results from some of our analyses but we need to work on them further before they are presentable, but things look promising! Data collection for this study will end by 31 July 2015 and we will be looking to publish some results over the next year. We have also started collecting data for another research study which aims to develop a protocol to assess the effects of treatment on abnormal brain networks in mental disorders. For this study, we are using a single dose of the memory enhancing agent galantamine and matched placebo. Healthy volunteers will be tested on two days and we will compare patterns of brain activity during task performance and at rest, and neurochemicals in key brain network nodes, following the administration of galantamine with similar observations following administration of the matched placebo. Although the aim is to develop a protocol to assess effects of treatment in mental disorders, we need to understand how any given treatment affects the healthy brain first. We started data collection for this study in October 2014 and aim to finish recruitment in the next few months. Once this is done, I will spend some time analysing the data from this study. Potential research questions will include: how does galantamine affect key brain networks during tasks and at rest? Does galantamine administration lead to a change in the concentration of neurochemicals? This past year overall has involved a lot of learning, I have gained a much deeper understanding of analysis techniques that I already had some experience with and I have also had the chance to learn new methods and skills. I am looking forward to polishing and fine-tuning all the work we've done so far and publish some papers, which I hope will happen before it's time to write my piece for the next annual review. J"

Schizophrenia International Research Conference - Florence

In April 2014 Jyothika had a poster accepted for presentation at the Schizophrenia International Research Conference in Florence, Italy. MHRUK was pleased to make a contribution to her attendance at the conference.