Functional MRI

3.3.1. What Is Functional MRI?

Functional Magnetic Resonance Imaging (fMRI) is a form of Magnetic Resonance Imaging that capitalizes on the varying magnetic properties of oxygenated and deoxygenated blood flow to create a variance in signal intensity indicating brain activity. FMRI images can be obtained during an active task or when the participant is at rest to examine regional activity as well as functional connectivity, defined as the correlated activity between two regions. Study design and data analyses of these two methods are discussed in more detail below.

3.3.2. How Functional MRI Works

In fMRI, quantification of the blood-oxygenation level dependent (BOLD) signal is used to assess locally increased blood flow in areas of neuronal activation. Specifically, when a brain region is active, blood flows to the area leading to an increase in the concentration of oxyhemoglobin relative to deoxyhemoglobin (Ricker and Arenth, 2008). Oxyhemoglobin is diamagnetic and deoxyhemoglobin is paramagnetic; their changing concentrations during activation cause local magnetic field inhomogeneities that can be detected using T2* weighted scans (Matthews, 2008). The changes in signal intensity from deoxyhemoglobin rich blood to oxyhemoglobin rich blood allows contrast between active and inactive brain regions. This BOLD signal change is generally quite small, from 1 to 6% (Tong et al., 2016). While the BOLD response is the foundation of the fMRI signal, it would be reductive to say that there is a 1:1 relationship between BOLD and neuronal activation. It is important to distinguish that the BOLD response is not indicative of direct neuronal activity but rather the synaptic and dendritic electrical potentials (Matthews, 2008).

Task-Based fMRI. Task-based fMRI assesses regional activation in response to a specific stimulus or task condition. While in the scanner, the participant is given a task, which can involve responses to visual or auditory stimuli or elicit more complex cognitive processes (i.e., decision-making). Tasks are selected or designed to tap a specific cognitive, emotional, or motor response or to "activate" a specific brain region. Task-based fMRI analyses typically utilize subtraction methods– examination of the BOLD signal during the condition of interest as compared to a control or comparison condition. This subtraction procedure is necessary to determine which areas are specifically activated during the task condition. Thus, all tasks require inclusion of such a control or contrast condition using either a block or event-related design. In a block design, the participant is exposed to experimental stimulus trials presented in blocks that are interleaved with blocks of a baseline/control condition. The block types are then compared. In event-related designs, the two stimuli are mixed, and analytic methods are used separate the responses to their respective stimuli (Stamatakis et al., 2017). Both experimental methods have their place. Block designs have increased statistical power in analysis and are more useful for detecting minute differences in activation (Friston et al., 1999). Event-related designs are more useful for events that cannot be presented all at once but rather require trial-by-trial shifts in conditions.

Resting-State fMRI. The spontaneous BOLD signal fluctuations that underlie resting-state studies are measured fundamentally differently from the measurement of the BOLD signal in task-based studies. In task-based studies, many repetitions of a task are analyzed against a control condition in order to remove random neuronal noise (Stamatakis et al., 2017). In resting-state studies, the emphasis is on the random neuronal noise, and there is no task-evoked response (Fox and Raichle, 2007). Specifically, resting-state fMRI capitalizes on the fact that the BOLD signal demonstrates low frequency oscillations (.01- 0.15 Hz) throughout the brain even when

"at rest." These oscillations are referred to as spontaneous BOLD signal fluctuations and they utilize nearly 20% of the body's energy expenditure (Shulman et al., 2004). Thus, understanding what the brain is doing with that energy is an important step in elucidating brain function. Regions of the brain demonstrate BOLD signals that oscillate in tandem even in the absence of any complex task (Biswal et al., 1995). These correlations in spontaneous neuronal activity are indicative of functional connectivity, which can be defined as the temporal dependency between spatially remote neurophysiological events (Friston et al., 1993). It is crucial to note that while functional connectivity implies some level of structural connectivity between regions, it is not a measure of direct anatomical connections (Greicius et al., 2009).

During resting-state fMRI, participants typically lie in the MRI scanner while instructed to relax and remain passive for anywhere from 6 to 30 minutes (Fox and Raichle, 2007). However, there are many different conditions for "rest." Some paradigms have participants keep their eyes closed, while others ask the participant to stare passively at a fixed point or complete a continuous active task. More recently, participants have been scanned while watching brief movies, with the aim of reducing head motion and sleepiness (Vanderwal et al., 2015). This method has been shown to improve detection of interindividual differences in functional connectivity over traditional rest scans (Vanderwal et al., 2017). Overall, given the variability in conditions used to define a relaxed state, (Buckner et al., 2013) proposed that the resting-state be looked at as just another task-based state. They argue that resting-states have their own underlying cognitive processes, and the functional areas active during resting-state parallel those that are activated during internally directed mental tasks.

3.3.3. Data Processing

Task-Based fMRI. Here, we will briefly summarize the most common preprocessing steps that are taken prior to fMRI data analyses, but it is important to note that the exact procedures and order may differ depending on study needs. The first step in processing fMRI data is typically brain extraction to remove any non-brain structures from the image, which is followed by slice timing correction. Each slice in an fMRI scan is taken at a slightly different timepoint; slice timing correction adjusts the data so that the voxels all appear to have been acquired at the same time (Smith, 2001). Head motion can have a significant impact on fMRI data; thus, motion correction algorithms are used to mitigate this effect (Woolrich et al., 2001). The resulting images then undergo various smoothing and filtering procedures. Spatial smoothing is applied to each volume of the fMRI dataset to reduce noise without affecting the activation signal (Woolrich et al., 2001). Intensity normalization adjusts volume intensity so that all volumes have the same mean intensity. High and low frequency filters are then applied to each voxel's time series to remove noise from heart rate, breathing and normal resting-state activation (Smith, 2001). The next steps are registration and normalization, which are critical when comparing scans across subjects. Registration is the process by which the fMRI scan is aligned with the subject's own structural MRI scan and normalization is the process by which the fMRI scans are then mapped to a standard space so that image related information can be compared between individuals (Woolrich et al., 2001).

After the previous preprocessing steps, the images are ready to undergo statistical analysis to determine which voxels are activated. General linear modeling (GLM) is typically used to determine areas of statistically significant activation (Smith, 2001). The linear model can be thought of as an assumption of activation. It is based upon the "stimulus function" which is an on/off waveform that describes the timing of the stimulus. This stimulus function is then combined with the hemodynamic response function (HRF), which represents the shape of the

BOLD signal's typically response. The result is the linear model, which is a function that describes how the brain is expected to activate in response to the stimulus (Smith, 2001). This resulting linear model is then fit to each voxel in order to estimate how well the voxel's behavior fits the model. This "goodness of fit" is defined by a corresponding Z value. If the Z value is significantly different from zero, then it can be concluded that activation has occurred (Smith, 2001). This statistical modeling step outputs a significance value for each voxel that describes how well the model of activation fits the voxel's time data. The resulting statistical map assigns a significance threshold to every voxel. However, this imposes a problem. Because the brain is described by so many voxels, even a significance of $p < .01$ means that hundreds of voxels will be marked as activated by chance alone. To solve this problem, Gaussian random field theory (GRF) is used for voxel thresholding. This method analyzes a smaller number of statistically independent voxels and rather considers clusters of activated voxels, applying the significance threshold to clusters rather than individual voxels. This lowers the probability of receiving a false positive result (Smith, 2001). After thresholding, the last remaining step is visualizing the activated areas of the brain. This is done by rendering the activated voxels in a specific color range, so that it is easy to identify the extent to which certain regions are active (Woolrich et al., 2001).

***Resting-State fMRI*.** Resting-state fMRI relies on much of the same preprocessing steps as task-based fMRI. The brain image is extracted from the surrounding anatomy, slice-timing corrections are applied, motion is corrected, and the subject's brain is mapped to a standardized space. Motion correction is particularly essential in resting-state fMRI as even the smallest head movements have been shown to alter functional connections (Power et al., 2015). Additionally, while low frequency spontaneous neuronal activity is filtered out of task-based fMRI data, this activity is the focus of resting-state fMRI (Heuvel and Pol, 2010).

As indicated earlier, task-based fMRI analyses aim to examine areas of activation in response to a specific task or stimulus. Alternatively, resting-state fMRI analyses focus on the temporal correlations in spontaneous neural activation between voxels of anatomically separated regions. Several different model-dependent and model-free methods are used to determine these "functional connections" (Heuvel and Pol, 2010). Model-dependent methods are also known as seed-based methods. In these analyses, resting-state timeseries data from one region of the brain (the seed) is correlated with the time-series data of all other brain regions. The resulting output of correlations is called the functional connectivity map, and it describes the functional connections of the seed region (Biswal et al., 1995). This method is computationally simplistic, and results in straightforward, easily interpretable results. The functional connectivity map allows for a clear understanding of which regions have connectivity with your selected seed .

Model-free methods allow for the exploration of whole brain connectivity patterns. They are not constrained by a single starting seed. Instead, they examine patterns of connectivity across the brain. Independent component analysis (ICA), principal component analysis, and clustering, are all common methods used to analyze resting-state time series (Heuvel and Pol, 2010). These model-free methods search for patterns in resting-state correlations across the whole brain and have revealed a number of canonical functional brain networks such as the "default mode network" which is discussed below in more detail (Raichle et al., 2001). Other networks have also been identified including the salience, sensorimotor, dorsal attention, and ventral attention networks, to name a few (Damoiseaux et al., 2006). As a result, resting-state fMRI methods have informed neural network models of brain function that are not easily

obtained using task-based designs. Because of this, and the ease of acquiring resting-state images, the study of the brain in its resting-state has become an area of intense research.

3.3.4. Applications

Functional MRI has been used to identify underlying neural mechanisms of various psychological disorders, such as schizophrenia, bipolar disorder, anxiety, and post-traumatic stress disorder. Alterations in functional connectivity as assessed using resting-state fMRI and in neural activation elicited using task-based methods have also been observed in patients with MDD. In this brief overview, both of these methods will be discussed as they pertain to depression research.

***Case Example: Depression*.** Task-based fMRI studies using tasks involving reward, emotion, and memory processing have demonstrated altered function of regions throughout the brain in patients with MDD, or at risk for MDD. Due to its relevance to MDD symptoms, the focus in this brief section will be on alterations in amygdala activity during emotion processing tasks. Atypical emotion processing is a hallmark symptom of MDD (Beck, 2008) and has been proposed to be associated with altered function of the amygdala. For example, studies have found that individuals with depression exhibit increased amygdala responses to negative emotional stimuli, as compared to healthy controls (Peluso et al., 2009, Suslow et al., 2010). Further, this increased amygdala response appears to attenuate with antidepressant treatment (Sheline et al., 2001, Murphy et al., 2009). Using an implicit emotion identification task, Arnone and colleagues (2012) found that after eight weeks of citalopram, the MDD patients who reached full remission showed significant decreases in both left and right amygdala responses to sad faces. No differences were observed between medicated and nonmedicated patients in remission. Taken together, these results underscore the complex role of the amygdala in emotion processing in individuals with MDD and suggest that remission of symptoms is associated with a reduction of amygdala hyper-reactivity whether due to the result of antidepressant treatment or not.

Resting-state fMRI has been used to identify differences in functional connectivity of brain regions and networks in patients with MDD. While, depression has been associated with atypical functional connectivity of several large-scale neural networks, here, we will focus on the default mode network (DMN). The DMN consists of the medial prefrontal cortex (mPFC), inferior parietal cortex, posterior cingulate cortex (PCC), and the precuneus (Raichle et al., 2001, Shulman et al., 1997), which have been identified to work together as a unified network based on resting-state findings. The DMN is implicated in different aspects of cognition, such as social understanding and self-awareness (Greicius et al., 2003, Li et al., 2014), in addition to rumination and self-referential processes, both of which are associated with key features of depression (Nolen-Hoeksema et al., 2008). In a meta-analysis, (Kaiser et al., 2015) Kaiser and colleagues (2015) found that MDD patients exhibited hyperconnectivity between DMN regions, the medial prefrontal cortex, hippocampus, and dorsal lateral prefrontal cortex, as compared to healthy controls. Further, stronger resting-state functional connectivity within the DMN has been associated with greater rumination and self-referencing in individuals with depression (Berman et al., 2014), which further supports studies identifying negative rumination as a key feature of depression (Holtzheimer and Mayberg, 2011). Resting-state fMRI has also been a useful tool for uncovering putative neural mechanisms of MDD treatments. For example, Liston and colleagues (2014) used resting-state fMRI to analyze between and within connectivity of the DMN and central executive network (CEN) in depressed patients before and after a five-week treatment of transcranial magnetic stimulation (TMS) of the dorsolateral prefrontal cortex (DLPFC), and healthy control subjects. At baseline, depressed patients exhibited greater functional connectivity

of the DMN than healthy controls. Following TMS, hyperconnectivity within the DMN was significantly reduced between the ventromedial prefrontal cortex, pregenual anterior cingulate cortex, and precuneus. This treatment effect was unique to the DMN and was not observed for the CEN, suggesting that changes in the functional connectivity of the DMN may be a mechanism of action of TMS treatment.

In summary, as these studies illustrate, both task-based and resting-state fMRI are extremely valuable to clinical research. While this section focused on depression, these imaging methods are useful for investigating many aspects of different types of psychopathology and have implications for both the understanding of underlying mechanisms of these disorders and measuring treatment effectiveness.

3.3.5. Advantages and Disadvantages

Advantages. Functional MRI has great utility for examining brain function. As it relies on the hydrogen atoms native to the body, and the hemodynamics of neuronal activation, it requires no exogenous tracers. Under high strength magnetic fields (greater than 1.5 Tesla), fMRI has greater spatial resolution than that of PET or SPECT. In higher strength magnetic fields (greater than 4 Tesla) BOLD signal changes can be detected from micro vessels and capillaries, and not just in veins and large venules (Ricker and Arenth, 2008). The magnetic fields utilized by MRI also do not carry the same risk of radiation that PET, SPECT, and CT scans incur (Gerber and Gonzalez, 2013). Furthermore, the elucidation of connected brain networks through non-invasive imaging is incredibly promising.

Disadvantages. Functional MRI has some drawbacks depending on the subject population and task at hand. Subjects must remain incredibly still, as slight movements cause artifacts in the imaging, and can even disturb the magnetic field (Ricker and Arenth, 2008). In resting-state studies, head motion has been linked to diminished connectivity between distant functional regions (Van Dijk et al., 2012). Even mouth movements used for conversational speech are too overt for fMRI. Thus, tasks must be designed to minimize movement, which, at times, also reduces their ecological validity and utility. Also, due to the high frequency noise of the scanner, the loud abrupt sounds of the magnetic coils, and the enclosed space of the machine, the experience may be aversive to potential participants (Ricker and Arenth, 2008).

3.3. Diffusion Tensor Imaging (DTI)

3.3.1. What Is DTI?

Diffusion Tensor Imaging (DTI) is a magnetic resonance-based technique that analyzes anatomic connectivity in the brain. It allows for a detailed evaluation of white matter structure and pathing. When used in conjunction with structural MRI data, DTI provides an important avenue for discerning both structure and organization of the brain and its constituent white matter. The ability to see physiological correlations in white matter tractography also allows for improved interpretation of functional MRI data. For example, DTI is often used to corroborate the anatomical connectivity of networks observed using resting-state fMRI functional connectivity methods (De Luca et al., 2006). As such, DTI has become a necessary step in bridging the gap between functional and structural imagery.

3.3.2. How DTI Works

In order to understand DTI, the underlying concepts must first be understood. The chief amongst them is anisotropy. Anisotropy describes the uneven motion of particles that DTI capitalizes on in order to measure microstructure and directionality of brain tissue, chiefly white matter. Normally, water’s motion is isotropic, meaning it moves equally in all directions.

However, within bodily tissue, the motion is obstructed by biological structures (Hagmann et al., 2006). Macromolecules and cell walls prevent the even distribution of motion. In the brain, the movement and directionality of water movement is usually relegated to tracts created by white matter axons. Diffusion occurs along the length of the axon within the cytoplasm, as water cannot easily diffuse into or out of the cell membrane. The degree to which anisotropy occurs is informative of the structure of the white matter. White matter that is more heavily myelinated has a thicker wall of tissue preventing the internal water molecules from diffusing out of the axon. While myelination prevents water from diffusing laterally out of the cell, it still allows for movement along the path of the axon (Wilde et al., 2017).

In order to quantify the anisotropic diffusion of water molecules and generate meaningful data, DTI relies on monitoring the activity of water molecules over time. Specifically, DTI measures the energy of water molecules in the brain at two time points. At the first time point, energy is imparted into the water molecules in the brain, and an MR image is made. This is called the B_0 time point and indicates the initial amount of water. A second image is taken 1000 milliseconds after. These sequential images become the basis for DTI image acquisition. If the water has diffused in the elapsed time period, the second image will indicate a lesser signal resulting from water content. (Hagmann et al., 2006). Therefore, there is an inverse relationship between water signal strength and the degree of water diffusion. To study the directionality of diffusion, water molecule movement in DTI is modeled along the x, y and z axes. By considering the movement along all axes, diffusion can be estimated in an ellipsoidal pattern, with the magnitude of motion being related to the length of the axes that create the ellipsoid. The longest axis of the ellipsoid indicates the greatest direction of motion, and typically lies along the axon of the neuron (Wilde et al., 2017). Therefore, an ellipsoid that has a longer, more extended shape represents a more anisotropic diffusion pattern, and a more spherical ellipsoid indicates more isotropic diffusion patterns (Basser, 2002).

The diffusion ellipsoid can be mathematically modeled as a 3x3 symmetric matrix, or a tensor (Soares et al., 2013). For our purposes, a tensor can be thought of as an array of values that can be solved to identify the magnitude and direction of diffusion. In order to fully define the tensor, a minimum of six diffusion directions must be measured (Le Bihan et al., 2001). However, sampling more diffusion directions results in a higher resolution image with fewer errors. For tracts of complex white matter modeling, it is not uncommon to take more than six measurements, and studies have used up to 64 diffusion directions (Alexander et al., 2007). While the diffusion ellipsoid is useful in understanding diffusion across multiple axes, it is not particularly effective for visualizing diffusion throughout the brain. To do this, tractography is used to reconstruct white matter tracts (Hagmann et al., 2006). Tractography involves visualization of white matter pathways using colors that represent the principal direction of diffusion.

3.3.3. Data Processing

DTI data can be analyzed in multiple ways. Two of the most common analyses are tractography and region of interest analysis, which rely on several measures, including fractional anisotropy, mean diffusivity, axial diffusivity, and radial diffusivity. Fractional anisotropy (FA) is a numerical value between 0 and 1 that represents the variance in diffusion along the main direction of motion. A value close to 1 indicates that the behavior is anisotropic, a value close to 0 is more isotropic. Greater FA indicates greater structural integrity of the white matter pathway (Soares et al., 2013). Axial diffusivity (AD) is the coefficient of diffusion across the long axis of the ellipsoid, typically lying along the axon. Radial (RD) diffusivity is the coefficient of

diffusion perpendicular to the long axis. Decreased axial diffusivity (AD), decreased fractional anisotropy (FA), and increased radial diffusivity (RD) are all indications of decreased white matter integrity (Hagmann et al., 2006). Mean diffusivity (MD) is the average of the magnitudes of diffusion across all three axes. These diffusion measurements (AD, RD and MD) are measured by the changes of water concentration following initial the B_0 time point.

Tractography is a DTI analysis technique that estimates the white matter pathways in the brain. It does this by looking at the continuity of diffusion measures along a vector. This indicates that there is a trajectory of water running through the vectors. This trajectory is called a "streamline." While streamlines are indicative of white matter pathways, they are not direct images, but rather mathematical implications (Wilde et al., 2017). The advantage of the DTI model is that it allows for a good determination of pathway orientation, as it can determine the direction and magnitude of diffusion. While conventional DTI has issues with resolving multiple tracts of intersecting white matter within a single voxel, new approaches to diffusion measurement have allowed for the resolving of crossing fibers (Wedeen et al., 2008). Traditional DTI relies on the assumption of Gaussian diffusion, with only a single diffusion tensor per voxel (Basser, 2002). However, mathematical models have been developed to incorporate non-Gaussian distributions and resolve multiple diffusion tensors within a single voxel (Madden et al., 2012).

For region of interest analyses or investigations of specific white matter tracts (Wilde et al., 2017), only one specific region of the brain is imaged, and the DTI measures for the area are calculated. This can be done in any selected area of the brain and is useful for determining the physiological effects of mental illnesses (Wilde et al., 2017, Snook et al., 2007). The technique is advantageous as it is less technically demanding and is more sensitive to smaller changes in structure (Park et al., 2004). However, ROI DTI analysis often involves co-registration of different sets of data, to consolidate into one coordinate plane. This can result in spatial distortion, which is compounded by the lower spatial resolution of DTI.

3.3.4. Applications

DTI has been commonly utilized in studies of aging and brain development and is an effective clinical tool used for diagnoses, analyzing the development of the brain in normal aging and neurodegeneration (Wilde et al., 2017). Here, we discuss MDD research studies that have used DTI to interrogate white matter integrity.

Case Example: Depression. As indicated earlier, MDD has been associated with alterations in functional brain networks such as the default mode network. DTI allows for researchers to determine whether alterations in functional networks extend to the structural connectivity of the brain. For example, in a whole brain DTI study of 95 MDD patients compared to 102 healthy controls, there were significant white matter connectivity differences in two distinct networks. Patients with MDD showed reduced white matter connectivity and integrity in regions of the default mode network, and a frontal-subcortical network that included medial orbitofrontal cortex (mOFC) and caudate regions (Korgaonkar et al., 2014a).

This finding of decreased white matter integrity has been demonstrated across multiple studies. Meta-analyses find evidence of decreased fractional anisotropy within the bilateral frontal cortex of depressed patients, an area that is commonly shown to be reduced in volume in structural MR studies (Liao et al., 2013, Kumar et al., 2000). However, the extent to which these white matter structural differences are a result of disease pathology or the effects of medication is unclear. A meta-analysis of microstructural brain abnormalities in unmedicated MDD participants found four regions of decreased fractional anisotropy, the right cerebellum

hemispheric lobule, the corpus callosum, and the bilateral superior longitudinal fasciculus (SLF) (Jiang et al., 2017). Subgroup analysis from this study indicated that participants suffering from MDD in medication washout studies and first episode medication naïve MDD participants showed different regional patterns of decreased FA. This finding suggests that either longer MDD duration, or previous medication, contributes to increased frontostriatal degeneration, which was not observed in the first episode patients.

DTI studies have recently been used to find predictors of treatment outcomes in patients suffering from MDD. In a 2014 study, DTI was used to identify predictive biomarkers within the white matter tracts of MDD patients. For example, FA of the stria terminalis, a key area of the limbic system, and the cingulate gyrus part of the cingulum have been shown to predict remission in patients receiving antidepressants (Korgaonkar et al., 2014b). The fractional anisotropy of the two tracts were 62% accurate at predicting remission, but accuracy increased to 74% when adding patient age to the model. A more recent DTI study found that MDD patients who responded positively to medication had greater white matter integrity than non-responders in several key regions of interest in the frontotemporal white matter and in the cortical systems including those involved in psychomotor and reward processes (Davis et al., 2019).

In summary, DTI studies have indicated it is possible to find biomarkers that are predictive of treatment outcomes. By analyzing key white matter pathways, researchers can determine whether an individual will respond well to SSRI medication. Whether it is by analyzing the white matter structures responsible for psychomotor and reward networks, or by the fractional anisotropy measures of the limbic system, it is clear that MDD treatments leave their mark on the structure of the brain (Davis et al., 2019, Jiang et al., 2017). These studies show that medication options result in lasting structural changes to the brain of MDD patients, and that these differences depend on the therapeutic response.

3.3.5. Advantages and Disadvantages

***Advantages*.** DTI provides detailed insight into structural neural pathways and networks, unique from any other neuroimaging modality. Prior to the invention of DTI, it was only possible to obtain measurements of white matter structure post-mortem (Mori and Zhang, 2006). Additional advantages of DTI include its brief acquisition time, accessibility and relevancy to clinical research, and availability for implementation (Descoteaux, 1999).

***Disadvantages*.** While DTI has many advantages, there are limitations that should also be considered. Due to its reliance on magnetic resonance imaging, DTI has the same disadvantages of structural MRI that have been previously discussed. In addition to this, DTI also requires a relatively complex processing pipeline above and beyond that needed for structural MRI; therefore, it does present unique disadvantages. One of the challenges of DTI is interpretation of the data, especially in a clinical context (O'Donnell and Westin, 2011). For example, many factors can influence changes in fractional anisotropy, such as cell death or changes in myelination (O'Donnell and Westin, 2011).

3.4.Magnetic Resonance Spectroscopy (MRS)

3.4.1. What Is MRS?

Magnetic Resonance Spectroscopy (MRS) relies on the same principles of magnetic resonance that MRI utilizes. However, while MRI utilizes resonance signals from hydrogen protons to generate an image of brain structure, MRS uses resonance signals from hydrogen and other atoms to produce spectral data representing chemical composition of molecules in the brain (Tognarelli et al., 2015). As MRS can be performed with most modern MRI machines with only

a few modifications, it is often added to scanning sequences in order to monitor brain biochemistry and metabolism. This allows for the creation of a neurochemical profile of a brain region in conjunction with the structure elucidated by MRI. Before MRS, there were no noninvasive techniques to assay the products of gene expression or metabolism in the human brain. In MRS, neuroscientists have a powerful technique that can work alongside other imaging methods to elucidate the internal biochemistry of the brain.

3.4.2. How MRS Works

There are many MRS methods available to clinicians and researchers that allow for the assessment of various neurometabolites. Initially, only ^{1}H MRS, also known as proton MRS, was used to non-invasively examine brain metabolism. However, as localization techniques advanced, ^{1}H MRS was joined by localized carbon-13, nitrogen-15, flourine-19, sodium-23, and phosphorus-31 spectroscopy (Gujar et al., 2005). Each targeted isotope comes with its own suite of pros and cons, and can be used to track specific metabolites in the brain. Currently, ^{1}H MRS is the most widely used MRS method in neurologic and psychiatric research. It utilizes the same radiofrequency waves as routine MRI scanning and therefore requires no upgrades or changes to the MRI machine, which makes it a cost-effective add-on to a scan. ^{1}H MRS is capable of recognizing 15 of the 80 brain metabolites observable through MRS (Lin et al., 2005), which include lipids, lactates, glutamate, creatine, choline, and myoinositol (Gujar et al., 2005). Spectroscopy utilizing the other isotopes can detect the presence of the other remaining metabolites, like transaminase and urea (Ross and Bluml, 2001).

MRS relies on three primary mechanisms of magnetic resonance to create a spectrum: nuclear spin, chemical shift, and spin – spin coupling (Tran et al., 2009). Nuclear spin is a property of atoms with magnetic nuclei, such as hydrogen and carbon-13. As described at the beginning of the MRI Methods section, an atom's spin can be thought of as the nucleus of an atom spinning about its axis. This spin creates a directional magnetic field. In the presence of a strong magnetic field, such as the one present in an MRI machine, the magnetic atoms become very organized, with some orienting in the same direction as the magnetic field (B_0), and some in the opposite direction (Tognarelli et al., 2015). When a radiofrequency of the appropriate amount of energy is applied it can flip the atoms that aligned with the B_0 field to the opposite direction; this is called resonance. When this occurs, a peak is created in the MRS spectrum. This phenomenon forms the basis of spectroscopy and allows for the determination of chemical structure. Molecules of different composition go into resonance and create peaks at different radiofrequencies. Examining the frequency at which a peak occurs offers clues to the chemical composition of a molecules. The area underneath the peak (or the integration) offers an indication of that molecule's relative concentration.

Atoms experience different magnetic fields depending upon their surrounding chemical environment. The electrons and dipole moments of neighboring atoms influence the overall applied field and the radiofrequency at which a specific atom goes into resonance and creates a peak in the MRS spectrum. This effect is called the chemical shift. Nuclei can be shielded from the applied magnetic field by a surrounding electron cloud or can be de-shielded by strongly positive magnetic fields (Tognarelli et al., 2015). This influences the radiofrequency at which a peak is created in the MRS spectrum. One can determine the composition of biological tissue by examining the positions of the peaks, or chemical shifts, of an MRS spectra resulting from this shielding. Specific molecules can be identified by their known chemical shift frequencies.

Spin-spin coupling is the phenomenon by which the magnetic spin of one atom influences the spin of a neighboring atom. This causes the spectra from the atoms to be split,

with multiple peaks centered around a central frequency. Depending on the number of peaks in the spectra, it is possible to determine the number and orientation of the adjacent atoms (Lin et al., 2005). The splitting pattern of the peaks provides information about the structure and topography of the molecule.

3.4.3. Data Processing

As discussed earlier, MRS is used to quantify neurochemicals throughout the brain based on the generated spectrum. As one of the most common applications in psychiatric research is examination of glutamate signaling, this will be discussed here as examples of how MRS data are processed and analyzed. There are three key features of an MRS spectrum that are analyzed to deduce chemical composition. The first feature is the position of the peaks, or the chemical shifts, which are caused by the different magnetic fields experienced by each atom in the scanned area. Molecules have characteristic chemical shifts, and so they can be used to determine identity. Chemical shift is described in units of parts per million (ppm) and is the x-axis of an MRS spectrum. In ^{1}H MRS, glutamate and its metabolite glutamine (collectively labeled Glx) create peaks in the MR spectra between 2.1 and 2.4 ppm (Lin et al., 2005). The second key feature is the integrated intensity, or the area under the peak of each signal. The integration provides information about the relative concentration of the molecules with larger concentrations being represented by peaks with larger integration. For example, the normal cerebral concentration of glutamate is between 3.0 and 12.5 mmol/kg (Tran et al., 2009). This characteristic of integration forms the basis for quantitative MRS, which is a technique that allows for the estimation of metabolite concentration within the brain (Alger, 2010). The last feature of the MRS spectrum is the spin-spin coupling pattern which manifests itself as multiple peaks centered about a central chemical shift. The pattern of the peaks provides additional information as to the structure of the molecule. For example, in an MRS spectrum with a Glx signal, the spin-spin coupling pattern manifests itself as two clusters of peaks between 2 and 2.5 ppm. One of the peaks is a triplet pattern, and the other is a quartet. Molecules that are structurally similar in turn have similar spin-spin coupling patterns (Bertholdo et al., 2013). Glutamate and glutamine are typically indistinguishable as a single signal in the ^{1}H MRS spectra, but in higher field strengths (>3T) it is possible to resolve the two peaks (Mullins et al., 2008).

3.4.4. Applications

MRS allows researchers and clinicians to track the presence of specific neurotransmitters throughout the brain in real time, making it an invaluable tool in neurological diagnoses and brain metabolism monitoring. A wide array of illnesses from Alzheimer's to hypoxia all have physiological indicators that can be observed through ^{1}H MRS (Tran et al., 2009). For example, glutamate can be a useful biomarker for stroke, lymphoma, and other metabolic brain disorders and the choline to creatine concentration ratio is a common indicator used by radiologists for tumor diagnoses (Lin et al., 2005). GABA and glutamate ^{1}H MRS are important tools for psychopharmacological inquiry. As GABA is one of the primary inhibitory neurotransmitters within the brain, and glutamate is the major excitatory neurotransmitter, drugs that regulate their concentration and transmission are of great clinical interest.

Case Example: Depression. MRS provides an invaluable tool when examining the neurobiological correlates of MDD, as it allows for the noninvasive observation of Glx metabolites and GABA concentrations. GABA and glutamate dysfunctions have long been implicated as a possible physiological cause for depressive symptoms (Duman et al., 2019). ^{1}H MRS has been pivotal in elucidating the role of GABAergic and glutamatergic neurometabolism in the disorder's pathology.

Preclinical rodent and primate studies of depression suggest significant neuronal atrophy in the hippocampus and prefrontal cortex hypothesized to result from the degeneration of excitatory glutamate projection neurons (Duman et al., 2016). ^{1}H MRS studies have been used to test this hypothesis in vivo in human patients. For example, researchers utilizing MRS have found lowered levels of glutamate neurometabolites in regions of the medial prefrontal cortex in unmedicated patients with MDD (Moriguchi et al., 2019). Furthermore, in conjunction with fMRI studies, decreased glutamate in the anterior cingulate cortex (ACC) is associated with decreased BOLD response to emotional stimuli in those with MDD (Lener et al., 2017).

Disruption of GABA modulation is theorized to reduce the integrity and control of excitatory neurotransmission in patients with MDD (Fee et al., 2017). GABA MRS studies have reported lower levels of GABA levels in cortical regions of patients suffering from MDD (Godfrey et al., 2018). This is consistent with postmortem studies of MDD patients that show decreased levels of the GABA synthetic enzyme GAD67 in the prefrontal cortex, and losses of GABAergic interneurons in the dorsolateral prefrontal cortex (Guilloux et al., 2012, Rajkowska et al., 2007). While GABA MRS studies show lower concentrations of the neurotransmitter in MDD patients, it appears to be state dependent, as patients in remission show no difference in GABA concentration when compared to non- depressed controls (Gerard Sanacora et al., 2003).

MRS techniques may be most promising in pharmacological studies of MDD. In the development of novel drugs, MRS is used to determine the degree to which the drug modulates the targeted neurometabolite, which allows for the expedited screening of promising candidates (Egerton, 2019). The clinical efficacy of drug treatments can also be examined using GABA and Glx concentrations as indicators of predicted therapeutic outcome (Egerton, 2019). This use of MRS to further develop and study drug mechanism has been used recently in the development of ketamine as a pharmacological treatment for MDD (Bojesen et al., 2018, Javitt et al., 2018). In studies of animal models of depression, ketamine-induced normalization of glutamate levels decreases regional cerebral blood flow (rCBF), which in turn prevents structural changes in the hippocampus (Schobel et al., 2013). An MRS study of healthy humans confirmed that a similar modulation of rCBF by the normalization of glutamate levels occurs in the anterior cingulate cortex after dosage with ketamine (Bojesen et al., 2018). This supports the use of ketamine to prevent MDD-related structural changes in the brain through the modulation of rCBF by glutamate management. Other studies of ketamine utilize MRS examinations of glutamate changes in the frontal and occipital lobe to understand the drug's therapeutic mechanism. In a study using 7T ^{1}H-MRS scanning, Evans et al., (2018) (Evans et al., 2018) found that contrary to expectations, there were no significant differences in metabolite concentrations between patients with MDD and healthy controls, or correlations between glutamate concentrations and mood. However, MDD subjects tended to have higher levels of glutamate concentration following ketamine infusions, indicating a greater sensitivity to the drug. Furthermore, there were two distinct groups of MDD patients: one that tended towards increases in glutamate concentration following ketamine infusion, and a second group that did not experience any increase . In short, MRS allows pharmaceutical researchers to examine a drug's direct effect on a specific metabolite, more quickly screen drug candidates, and then translate findings from rodent to humans using the same biomarker.

3.4.5. Advantages and Disadvantages

Advantages. MRS is largely available in any facility with a modern MRI scanner, often allowing it to be relatively inexpensive and accessible to add on to MRI scanning sequences . Further, it can complement other functional and structural neuroimaging techniques by providing

unique information concerning the chemical composition of the scanned brain area. In addition, in vivo MRS is non-invasive, and does not expose subjects to ionizing radiation as methods such as PET and SPECT do (Gerber and Gonzalez, 2013).

Disadvantages. MRS has the same contraindications as other MRI-based methods. In addition, compared to other MRI-based methods, in vivo MRS has relatively low spatial and temporal resolution, as well as a low signal-to-noise ratio (Gerber and Gonzalez, 2013, Tognarelli et al., 2015, Alger, 2010). Furthermore, the general limited ability of MRS to build images causes precise visual interpretation of results to be difficult. Quantitative MRS measures of metabolite concentrations are associated with potential challenges as well. For example, methods involving quantification relative to another metabolite may require certain assumptions about the system. For instance, one might assume that creatine levels do not change across healthy and diseased states when using creatine as a reference metabolite. Although absolute quantification appears theoretically more desirable, it is often difficult practically, also involves using ratios (with a different reference signal), and can be biased by methodological choices and assumptions as well. True metabolite concentrations are being estimated, and precision greatly depends on the methodology used. Overall, reliable MRS quantification is quite complex, but is possible when one does account for the many considerations that are required for accuracy (Alger, 2010).

Conclusion

In summary, the goal of this chapter was to summarize neuroimaging methods in order to make them more accessible to the broader clinical psychology community. While each technique comes with its own advantages and disadvantages, they have all contributed significantly to current neurobiological models of psychopathology, including major depression as detailed in this chapter. Additionally, they have contributed to the development of pharmacological interventions and to our understanding of underlying mechanisms of effective psychotherapy. As technological advances continue to be made, such methods will be further refined are sure to lead to the development of multiple methods for studying the human brain *in vivo*.

References

ALEXANDER, A. L., LEE, J. E., LAZAR, M. & FIELD, A. S. 2007. Diffusion tensor imaging of the brain. *Neurotherapeutics : the journal of the American Society for Experimental NeuroTherapeutics,* 4**,** 316-329.

ALGER, J. R. 2010. Quantitative proton magnetic resonance spectroscopy and spectroscopic imaging of the brain: a didactic review. *Top Magn Reson Imaging,* 21**,** 115-28.

ARNONE, D., MCKIE, S., ELLIOTT, R., THOMAS, E. J., DOWNEY, D., JUHASZ, G., WILLIAMS, S. R., DEAKIN, J. F. & ANDERSON, I. M. 2012. Increased amygdala responses to sad but not fearful faces in major depression: relation to mood state and pharmacological treatment. *Am J Psychiatry,* 169**,** 841-50.

BASSER, P. J. 2002. Relationships between diffusion tensor and q-space MRI. *Magn Reson Med,* 47**,** 392-7.

BECK, A. T. 2008. The evolution of the cognitive model of depression and its neurobiological correlates. *Am J Psychiatry,* 165**,** 969-77.

BERMAN, M. G., MISIC, B., BUSCHKUEHL, M., KROSS, E., DELDIN, P. J., PELTIER, S., CHURCHILL, N. W., JAEGGI, S. M., VAKORIN, V., MCINTOSH, A. R. & JONIDES, J. 2014. Does resting-state connectivity reflect depressive rumination? A tale of two analyses. *Neuroimage,* 103**,** 267-279.

BERTHOLDO, D., WATCHARAKORN, A. & CASTILLO, M. 2013. Brain proton magnetic resonance spectroscopy: introduction and overview. *Neuroimaging clinics of North America,* 23**,** 359-380.

BISWAL, B., ZERRIN YETKIN, F., HAUGHTON, V. M. & HYDE, J. S. 1995. Functional connectivity in the motor cortex of resting human brain using echo-planar mri. *Magnetic Resonance in Medicine,* 34**,** 537-541.

BOJESEN, K. B., ANDERSEN, K. A., RASMUSSEN, S. N., BAANDRUP, L., MADSEN, L. M., GLENTHØJ, B. Y., ROSTRUP, E. & BROBERG, B. V. 2018. Glutamate Levels and Resting Cerebral Blood Flow in Anterior Cingulate Cortex Are Associated at Rest and Immediately Following Infusion of S-Ketamine in Healthy Volunteers. *Frontiers in Psychiatry,* 9.

BUCKNER, R. L., KRIENEN, F. M. & YEO, B. T. 2013. Opportunities and limitations of intrinsic functional connectivity MRI. *Nat Neurosci,* 16**,** 832-7.

DAMOISEAUX, J. S., ROMBOUTS, S. A. R. B., BARKHOF, F., SCHELTENS, P., STAM, C. J., SMITH, S. M. & BECKMANN, C. F. 2006. Consistent resting-state networks across healthy subjects. *Proceedings of the National Academy of Sciences,* 103**,** 13848-13853.

DAVIS, A. D., HASSEL, S., ARNOTT, S. R., HARRIS, J., LAM, R. W., MILEV, R., ROTZINGER, S., ZAMYADI, M., FREY, B. N., MINUZZI, L., STROTHER, S. C., MACQUEEN, G. M., KENNEDY, S. H. & HALL, G. B. 2019. White Matter Indices of Medication Response in Major Depression: A Diffusion Tensor Imaging Study. *Biological Psychiatry: Cognitive Neuroscience and Neuroimaging,* 4**,** 913-924.

DE LUCA, M., BECKMANN, C. F., DE STEFANO, N., MATTHEWS, P. M. & SMITH, S. M. 2006. fMRI resting state networks define distinct modes of long-distance interactions in the human brain. *Neuroimage,* 29**,** 1359-67.

DESCOTEAUX, M. 1999. High Angular Resolution Diffusion Imaging (HARDI). *Wiley Encyclopedia of Electrical and Electronics Engineering.*

DUMAN, R. S., AGHAJANIAN, G. K., SANACORA, G. & KRYSTAL, J. H. 2016. Synaptic plasticity and depression: new insights from stress and rapid-acting antidepressants. *Nature Medicine,* 22**,** 238-249.

DUMAN, R. S., SANACORA, G. & KRYSTAL, J. H. 2019. Altered Connectivity in Depression: GABA and Glutamate Neurotransmitter Deficits and Reversal by Novel Treatments. *Neuron,* 102**,** 75-90.

EGERTON, A. 2019. The potential of 1H-MRS in CNS drug development. *Psychopharmacology*.

EVANS, J. W., LALLY, N., AN, L., LI, N., NUGENT, A. C., BANERJEE, D., SNIDER, S. L., SHEN, J., ROISER, J. P. & ZARATE, C. A., JR. 2018. 7T (1)H-MRS in major depressive disorder: a Ketamine Treatment Study. *Neuropsychopharmacology,* 43**,** 1908-1914.

FEE, C., BANASR, M. & SIBILLE, E. 2017. Somatostatin-Positive Gamma-Aminobutyric Acid Interneuron Deficits in Depression: Cortical Microcircuit and Therapeutic Perspectives. *Biol Psychiatry,* 82**,** 549-559.

FOX, M. D. & RAICHLE, M. E. 2007. Spontaneous fluctuations in brain activity observed with functional magnetic resonance imaging. *Nat Rev Neurosci,* 8**,** 700-11.

FRISTON, K. J., FRITH, C. D., LIDDLE, P. F. & FRACKOWIAK, R. S. J. 1993. Functional Connectivity: The Principal-Component Analysis of Large (PET) Data Sets. *Journal of Cerebral Blood Flow & Metabolism,* 13**,** 5-14.

FRISTON, K. J., ZARAHN, E., JOSEPHS, O., HENSON, R. N. A. & DALE, A. M. 1999. Stochastic Designs in Event-Related fMRI. *NeuroImage,* 10**,** 607-619.

GERARD SANACORA, M.D., PH.D. ,, GRAEME F. MASON, PH.D. ,, DOUGLAS L. ROTHMAN, PH.D. ,, FAHMEED HYDER, M.D. ,, JAMES J. CIARCIA, M.D. ,, ROBERT B. OSTROFF, M.D. ,, ROBERT M. BERMAN, M.D. , AND & JOHN H. KRYSTAL, M.D. 2003. Increased Cortical GABA Concentrations in Depressed Patients Receiving ECT. *American Journal of Psychiatry,* 160**,** 577-579.

GERBER, A. J. & GONZALEZ, M. Z. 2013. Structural and Functional Brain Imaging in Clinical Psychology. *In:* COMER, J. S., KENDALL, P.C. (ed.) *The Oxford Handbook of Research Strategies for Clinical Psychology.* New York, NY: Oxford University Press.

GODFREY, K. E. M., GARDNER, A. C., KWON, S., CHEA, W. & MUTHUKUMARASWAMY, S. D. 2018. Differences in excitatory and inhibitory neurotransmitter levels between depressed patients and healthy controls: A systematic review and meta-analysis. *Journal of Psychiatric Research,* 105**,** 33-44.

GREICIUS, M. D., KRASNOW, B., REISS, A. L. & MENON, V. 2003. Functional connectivity in the resting brain: a network analysis of the default mode hypothesis. *Proc Natl Acad Sci U S A,* 100**,** 253-8.

GREICIUS, M. D., SUPEKAR, K., MENON, V. & DOUGHERTY, R. F. 2009. Resting-state functional connectivity reflects structural connectivity in the default mode network. *Cereb Cortex,* 19**,** 72-8.

GUILLOUX, J. P., DOUILLARD-GUILLOUX, G., KOTA, R., WANG, X., GARDIER, A. M., MARTINOWICH, K., TSENG, G. C., LEWIS, D. A. & SIBILLE, E. 2012. Molecular evidence for BDNF- and GABA-related dysfunctions in the amygdala of female subjects with major depression. *Molecular Psychiatry,* 17**,** 1130-1142.

GUJAR, S. K., MAHESHWARI, S., BJÖRKMAN-BURTSCHER, I. & SUNDGREN, P. C. 2005. Magnetic resonance spectroscopy. *Journal of neuro-ophthalmology : the official journal of the North American Neuro-Ophthalmology Society,* 25**,** 217-226.

HAGMANN, P., JONASSON, L., MAEDER, P., THIRAN, J. P., WEDEEN, V. J. & MEULI, R. 2006. Understanding diffusion MR imaging techniques: from scalar diffusion-weighted imaging to diffusion tensor imaging and beyond. *Radiographics,* 26 Suppl 1**,** S205-23.

HEUVEL, M. & POL, H. 2010. Exploring the brain network: A review on resting-state fMRI functional connectivity. *European neuropsychopharmacology : the journal of the European College of Neuropsychopharmacology,* 20**,** 519-34.

HOLTZHEIMER, P. E. & MAYBERG, H. S. 2011. Stuck in a rut: rethinking depression and its treatment. *Trends Neurosci,* 34**,** 1-9.

JAVITT, D. C., CARTER, C. S., KRYSTAL, J. H., KANTROWITZ, J. T., GIRGIS, R. R., KEGELES, L. S., RAGLAND, J. D., MADDOCK, R. J., LESH, T. A., TANASE, C., CORLETT, P. R., ROTHMAN, D. L., MASON, G., QIU, M., ROBINSON, J., POTTER, W. Z., CARLSON, M., WALL, M. M., CHOO, T. H., GRINBAND, J. & LIEBERMAN, J. A. 2018. Utility of Imaging-Based Biomarkers for Glutamate-Targeted Drug Development in Psychotic Disorders: A Randomized Clinical Trial. *JAMA Psychiatry,* 75**,** 11-19.

JIANG, J., ZHAO, Y. J., HU, X. Y., DU, M. Y., CHEN, Z. Q., WU, M., LI, K. M., ZHU, H. Y., KUMAR, P. & GONG, Q. Y. 2017. Microstructural brain abnormalities in medication-free patients with major depressive disorder: a systematic review and meta-analysis of diffusion tensor imaging. *J Psychiatry Neurosci,* 42**,** 150-163.

KAISER, R. H., ANDREWS-HANNA, J. R., WAGER, T. D. & PIZZAGALLI, D. A. 2015. Large-Scale Network Dysfunction in Major Depressive Disorder: A Meta-analysis of Resting-State Functional Connectivity. *JAMA Psychiatry,* 72**,** 603-11.

KORGAONKAR, M. S., FORNITO, A., WILLIAMS, L. M. & GRIEVE, S. M. 2014a. Abnormal Structural Networks Characterize Major Depressive Disorder: A Connectome Analysis. *Biological Psychiatry,* 76**,** 567-574.

KORGAONKAR, M. S., WILLIAMS, L. M., SONG, Y. J., USHERWOOD, T. & GRIEVE, S. M. 2014b. Diffusion tensor imaging predictors of treatment outcomes in major depressive disorder. *British Journal of Psychiatry,* 205**,** 321-328.

LE BIHAN, D., MANGIN, J.-F., POUPON, C., CLARK, C. A., PAPPATA, S., MOLKO, N. & CHABRIAT, H. 2001. Diffusion tensor imaging: Concepts and applications. *Journal of Magnetic Resonance Imaging,* 13**,** 534-546.

LENER, M. S., NICIU, M. J., BALLARD, E. D., PARK, M., PARK, L. T., NUGENT, A. C. & ZARATE, C. A. 2017. Glutamate and Gamma-Aminobutyric Acid Systems in the Pathophysiology of Major Depression and Antidepressant Response to Ketamine. *Biological Psychiatry,* 81**,** 886-897.

LI, W., MAI, X. & LIU, C. 2014. The default mode network and social understanding of others: what do brain connectivity studies tell us. *Front Hum Neurosci,* 8**,** 74.

LIN, A., ROSS, B. D., HARRIS, K. & WONG, W. 2005. Efficacy of proton magnetic resonance spectroscopy in neurological diagnosis and neurotherapeutic decision making. *NeuroRX,* 2**,** 197-214.

MADDEN, D. J., BENNETT, I. J., BURZYNSKA, A., POTTER, G. G., CHEN, N.-K. & SONG, A. W. 2012. Diffusion tensor imaging of cerebral white matter integrity in cognitive aging. *Biochimica et biophysica acta,* 1822**,** 386-400.

MATTHEWS, P. M. 2008. An introduction to functional magnetic resonance imaging of the brain. *In:* JEZZARD, P., MATTHEWS, P. M. & SMITH, S. M. (eds.) *Functional MRI : an introduction to methods.* 1 ed.: Oxford University Press.

MORI, S. & ZHANG, J. 2006. Principles of diffusion tensor imaging and its applications to basic neuroscience research. *Neuron,* 51**,** 527-39.

MORIGUCHI, S., TAKAMIYA, A., NODA, Y., HORITA, N., WADA, M., TSUGAWA, S., PLITMAN, E., SANO, Y., TARUMI, R., ELSALHY, M., KATAYAMA, N., OGYU, K., MIYAZAKI, T., KISHIMOTO, T., GRAFF-GUERRERO, A., MEYER, J. H., BLUMBERGER, D. M., DASKALAKIS, Z. J., MIMURA, M. & NAKAJIMA, S. 2019. Glutamatergic neurometabolite levels in major depressive disorder: a systematic review and meta-analysis of proton magnetic resonance spectroscopy studies. *Molecular Psychiatry,* 24**,** 952-964.

MULLINS, P. G., CHEN, H., XU, J., CAPRIHAN, A. & GASPAROVIC, C. 2008. Comparative reliability of proton spectroscopy techniques designed to improve detection of J-coupled metabolites. *Magn Reson Med,* 60**,** 964-9.

MURPHY, S. E., NORBURY, R., O'SULLIVAN, U., COWEN, P. J. & HARMER, C. J. 2009. Effect of a single dose of citalopram on amygdala response to emotional faces. *Br J Psychiatry,* 194**,** 535-40.

NOLEN-HOEKSEMA, S., WISCO, B. E. & LYUBOMIRSKY, S. 2008. Rethinking Rumination. *Perspect Psychol Sci,* 3**,** 400-24.

O'DONNELL, L. J. & WESTIN, C. F. 2011. An introduction to diffusion tensor image analysis. *Neurosurg Clin N Am,* 22**,** 185-96, viii.

PARK, H.-J., WESTIN, C.-F., KUBICKI, M., MAIER, S. E., NIZNIKIEWICZ, M., BAER, A., FRUMIN, M., KIKINIS, R., JOLESZ, F. A., MCCARLEY, R. W. & SHENTON, M. E. 2004. White matter hemisphere asymmetries in healthy subjects and in schizophrenia: a diffusion tensor MRI study. *NeuroImage,* 23**,** 213-223.

PELUSO, M. A., GLAHN, D. C., MATSUO, K., MONKUL, E. S., NAJT, P., ZAMARRIPA, F., LI, J., LANCASTER, J. L., FOX, P. T., GAO, J. H. & SOARES, J. C. 2009. Amygdala hyperactivation in untreated depressed individuals. *Psychiatry Res,* 173**,** 158-61.

POWER, J. D., SCHLAGGAR, B. L. & PETERSEN, S. E. 2015. Recent progress and outstanding issues in motion correction in resting state fMRI. *Neuroimage,* 105**,** 536-51.

RAICHLE, M. E., MACLEOD, A. M., SNYDER, A. Z., POWERS, W. J., GUSNARD, D. A. & SHULMAN, G. L. 2001. A default mode of brain function. *Proc Natl Acad Sci U S A,* 98**,** 676-82.

RAJKOWSKA, G., O'DWYER, G., TELEKI, Z., STOCKMEIER, C. A. & MIGUEL-HIDALGO, J. J. 2007. GABAergic Neurons Immunoreactive for Calcium Binding Proteins are Reduced in the Prefrontal Cortex in Major Depression. *Neuropsychopharmacology,* 32**,** 471-482.

RICKER, J. H. & ARENTH, P. M. 2008. Functional neuroimaging in clinical neuropsychology. *In:* MORGAN, J. E. & RICKER, J. H. (eds.) *Textbook of clinical neuropsychology.* New York, NY: Psychology Press.

ROSS, B. & BLUML, S. 2001. Magnetic resonance spectroscopy of the human brain. *The Anatomical Record,* 265**,** 54-84.

SCHOBEL, SCOTT A., CHAUDHURY, NASHID H., KHAN, USMAN A., PANIAGUA, B., STYNER, MARTIN A., ASLLANI, I., INBAR, BENJAMIN P., CORCORAN, CHERYL M., LIEBERMAN, JEFFREY A., MOORE, H. & SMALL, SCOTT A. 2013. Imaging Patients with Psychosis and a Mouse Model Establishes a Spreading Pattern of Hippocampal Dysfunction and Implicates Glutamate as a Driver. *Neuron,* 78**,** 81-93.

SHELINE, Y. I., BARCH, D. M., DONNELLY, J. M., OLLINGER, J. M., SNYDER, A. Z. & MINTUN, M. A. 2001. Increased amygdala response to masked emotional faces in depressed subjects resolves with antidepressant treatment: an fMRI study. *Biol Psychiatry,* 50**,** 651-8.

SHULMAN, G. L., FIEZ, J. A., CORBETTA, M., BUCKNER, R. L., MIEZIN, F. M., RAICHLE, M. E. & PETERSEN, S. E. 1997. Common Blood Flow Changes across Visual Tasks: II. Decreases in Cerebral Cortex. *J Cogn Neurosci,* 9**,** 648-63.

SHULMAN, R. G., ROTHMAN, D. L., BEHAR, K. L. & HYDER, F. 2004. Energetic basis of brain activity: implications for neuroimaging. *Trends Neurosci,* 27**,** 489-95.

SMITH, S. M. 2001. Overview of fMRI analysis. *In:* PETER JEZZARD, P. M. M., STEPHEN M. SMITH (ed.) *Functional Magnetic Resonance Imaging: An Introduction to Methods 1st Edition.* 1 ed. New York, New York: Oxford University Press.

SNOOK, L., PLEWES, C. & BEAULIEU, C. 2007. Voxel based versus region of interest analysis in diffusion tensor imaging of neurodevelopment. *NeuroImage,* 34**,** 243-252.

SOARES, J. M., MARQUES, P., ALVES, V. & SOUSA, N. 2013. A hitchhiker's guide to diffusion tensor imaging. *Front Neurosci,* 7**,** 31.

STAMATAKIS, E. A., ORFANIDOU, E. & PAPANICOLAOU, A. C. 2017. Functional Magnetic Resonance Imaging. *In:* PAPANICOLAOU, A. C. (ed.) *The Oxford Handbook of Functional Brain Imaging in Neuropsychology and Cognitive Neurosciences.* New York, NY: Oxford University Press.

SUSLOW, T., KONRAD, C., KUGEL, H., RUMSTADT, D., ZWITSERLOOD, P., SCHONING, S., OHRMANN, P., BAUER, J., PYKA, M., KERSTING, A., AROLT, V., HEINDEL, W. & DANNLOWSKI, U. 2010. Automatic mood-congruent amygdala responses to masked facial expressions in major depression. *Biol Psychiatry,* 67**,** 155-60.

TOGNARELLI, J. M., DAWOOD, M., SHARIFF, M. I. F., GROVER, V. P. B., CROSSEY, M. M. E., COX, I. J., TAYLOR-ROBINSON, S. D. & MCPHAIL, M. J. W. 2015. Magnetic Resonance Spectroscopy: Principles and Techniques: Lessons for Clinicians. *Journal of Clinical and Experimental Hepatology,* 5**,** 320-328.

TONG, Y., HOCKE, L. M., LINDSEY, K. P., ERDOĞAN, S. B., VITALIANO, G., CAINE, C. E. & FREDERICK, B. D. 2016. Systemic Low-Frequency Oscillations in BOLD Signal Vary with Tissue Type. *Frontiers in Neuroscience,* 10.

TRAN, T., ROSS, B. & LIN, A. 2009. Magnetic Resonance Spectroscopy in Neurological Diagnosis. *Neurologic Clinics,* 27**,** 21-60.

VAN DIJK, K. R., SABUNCU, M. R. & BUCKNER, R. L. 2012. The influence of head motion on intrinsic functional connectivity MRI. *Neuroimage,* 59**,** 431-8.

VANDERWAL, T., EILBOTT, J., FINN, E. S., CRADDOCK, R. C., TURNBULL, A. & CASTELLANOS, F. X. 2017. Individual differences in functional connectivity during naturalistic viewing conditions. *Neuroimage,* 157**,** 521-530.

VANDERWAL, T., KELLY, C., EILBOTT, J., MAYES, L. C. & CASTELLANOS, F. X. 2015. Inscapes: A movie paradigm to improve compliance in functional magnetic resonance imaging. *NeuroImage,* 122**,** 222-232.

WEDEEN, V. J., WANG, R. P., SCHMAHMANN, J. D., BENNER, T., TSENG, W. Y., DAI, G., PANDYA, D. N., HAGMANN, P., D'ARCEUIL, H. & DE CRESPIGNY, A. J. 2008. Diffusion spectrum magnetic resonance imaging (DSI) tractography of crossing fibers. *Neuroimage,* 41**,** 1267-77.

WILDE, E. A., AYOUB, K. W. & CHOUDHRI, A. F. 2017. Diffusion Tensor Imaging: The Confluence of Structural and Functional Images. *In:* PAPANICOLAOU, A. C. (ed.) *The Oxford Handbook of Functional Brain Imaging in Neuropsychology and Cognitive Neurosciences.* New York, NY: Oxford University Press.

WOOLRICH, M. W., RIPLEY, B. D., BRADY, M. & SMITH, S. M. 2001. Temporal Autocorrelation in Univariate Linear Modeling of FMRI Data. *NeuroImage,* 14**,** 1370-1386.