CHAPTER 6

General discussion

Depressive symptoms in the general population are of clinical relevance as they increase the risk of developing a major depressive episode (MDE; Cuijpers and Smit, 2004; Karsten et al., 2011) which in turn increases the probability of another episode (Hardeveld et al., 2010). Therefore, preventing the onset of the first episode is crucial. But, what is it that makes some people more vulnerable to depression? Neuroimaging studies have long been suggesting major depression disorder (MDD) as a disorder of abnormal brain connectivity. In other words, neuronal communication seems to be impaired in individuals with depression. However, because the most common strategy used to understand the neuronal basis of depression is to focus on studying differences between healthy and patient populations, it is currently unclear whether depressive symptoms in individuals without an MDD diagnosis have reliable functional brain correlates. Accordingly, the first aim of the thesis was to investigate whether abnormalities in network connectivity can already be seen in the brain of healthy individuals who might be at higher risk of developing MDD. More specifically, in Chapter 2 we explored the overall dynamics of the brain and how this could be affected in undiagnosed individuals with current symptoms of depression. Additionally, we examined the specific orchestration of brain network interactions in the presence of depressive symptoms in Chapter 3.

The second aim of the thesis was to elucidate the neuronal mechanisms associated with a major depressive episode (MDE) beyond those implicated in MDD. To this end, in Chapter 4 we investigated whether and how the dynamic interactions of the brain change as depressed patients with current symptomatic remission experience a new depressive episode.

The third and final aim took a methodological direction to elucidate to what extent fluctuations in brain connectivity, as captured by fMRI, support the complexity inherent to brain communication. Specifically, in Chapter 5 we used task and rest functional MRI data from healthy individuals to characterize the dimensionality of brain connectivity fluctuations across three different experimental conditions.
The key findings of each chapter are briefly summarized below:

**Chapter 2** uncovers abnormal patterns of whole-brain connectivity associated with depressive symptoms in nonclinical individuals. The results show that the dynamical complexity that underlies brain connectivity gradually decreases as individuals report depressive symptoms more frequently. The frequency of the symptoms was found to be associated with a less hierarchical organization across brain regions and less variability of brain connectivity over time. These imbalances may be responsible for the observed reduced capacity of the brain to integrate information among people with elevated depressive symptoms.

**Chapter 3** reveals changes in brain network connectivity at the macroscopic level that are associated with the presence of depressive symptoms in nonclinical individuals. We found imbalances in the brain's dynamical repertoire, which is the dynamic configuration in which brain networks evolve and dissolve over time. Specifically, individuals with more frequent depressive symptoms engaged more in states connecting regions of the default mode, memory retrieval and frontoparietal network and less in states connecting mostly the visual and dorsal attention systems.

**Chapter 4** shows changes in the brain's dynamical repertoire as MDD patients in absence of symptomatology experience a new depressive episode. These changes were specifically seen in a state connecting frontoparietal areas with areas from the default mode network. This study provides evidence of alterations in the brain's dynamical repertoire that are specifically associated with the presence of depressive symptoms.

**Chapter 5** studies how and why the pattern of interactions between brain networks fluctuates over time. The analyzed data consisted of functional MRI from 100 healthy subjects across three different experimental conditions, which were obtained from the Human Connectome Project. This study demonstrates that modulations in neuronal communication, represented as changes in network interactions, fluctuate within a multidimensional manifold that can be reliably measured.

**6.2 Implications of the thesis findings**

The findings of this thesis have important implications in at least two major respects.

**6.2.1 Neurological relevance of depressive symptoms in the general population**

Depressive symptoms in the general population are often triggered by the experience of a stressful or upsetting life event (Kendler et al., 1999). In particular, the breakup of a relationship is often seen as a stressful situation that is accompanied by depression-like symptoms, such as sadness and general loss of interest (Field et al., 2009; Kendler et al., 1999; Najib et al., 2004). Although they tend to disappear within the early months after the event, for some people these symptoms persist (Field et al., 2009). Consequently, this has a negative impact on mental health and places the very individuals at higher risk of developing MDD (Cuijpers and Smit, 2004; Karsten et al., 2011; Zisook et al., 2010). This thesis set out to investigate depressive symptoms in the healthy population and the associated neural correlates as an initial step towards understanding depression vulnerability.

Accumulating evidence suggests that depressive symptoms may result from abnormal interactions within and between several brain networks (Wang et al., 2012; Kaiser et al., 2015; Mulders et al., 2015; Helm et al., 2018). More recently, it has been suggested that the pattern of transitions between networks could serve as a biomarker for MDD. These include aberrations in the dynamics involving the default mode (Wise et al., 2017), frontoparietal and salience (Demirtaş et al., 2016; Kaiser et al., 2015), and the cognitive control network (Figueroa et al., 2019). In Chapter 3, we found a default mode network dominance also in nonclinical individuals who reported frequent depressive symptoms. This dominance has been linked to an excessive self-referential thought and a tendency to ruminate (Broyd et al., 2009; Christoff et al., 2009; J. P. Hamilton et al., 2011; Marchetti et al., 2012; Marusak et al., 2017) — which have been considered a risk factor in the development and maintenance of mental disorders (Michalak et al., 2011).

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Depression has been linked predominantly to imbalances in whole-brain network interactions. These imbalances are thought to harm the functional organization of the brain, hampering effective cognition and healthy behavior. However, to the best
of our knowledge, no studies have investigated how the experience of depressive symptoms in nonclinical populations relates to changes in the brain’s global organization. As detailed in Chapter 2, we found that depressive symptoms in nonclinical subjects are associated with more rigid patterns of whole-brain functional connectivity. Furthermore, this lack of dynamism in the functional organization of the brain was accompanied by a reduction of the brain’s capacity to integrate and process the incoming flow of information across distributed regions. This inefficient broadcasting of information throughout the brain, resulting from the static ways in which brain regions communicate over time, may account for the maladaptive cognitive processes underlying depressive symptoms such as repetitive thinking and impaired inhibition (Nolen-hoeksema et al., 1993).

Altogether, our studies on depressive symptoms in nonclinical individuals suggest that imbalances in the complex dynamics of the brain may promote maladaptive coping strategies such as a repeated focusing on unpleasant thoughts, which inhibits engaging in more enjoyable thoughts. This tendency potentially puts healthy people at higher risk of developing depression, which accentuates the importance of investigating depressive symptoms in the general population.

### 6.2.2 Fluctuations in network connectivity contain depression-related information

It is clear now that the functional organization of the brain is not static. This means that the patterns of communication between brain regions do not follow rigid routes, rather dynamic trajectories of coordinated activity. Accumulating evidence indicates that functional connectivity derived from resting-state fMRI contains ample dynamic information (Calhoun et al., 2014; Global Burden of Disease Study 2013 Collaborators, 2015; Hutchison et al., 2013). However, whether fMRI functional connectivity enables to reliably capture meaningful fluctuations remains a challenging research topic (Calhoun et al., 2014; Lurie et al., 2020). In Chapter 5, we demonstrated that configurations of functional connectivity fluctuate within a multidimensional manifold that can be reliably measured. Along these dimensions, brain regions are thought to interact in a flexible and coordinated manner to allow the integration of information (e.g., sensory input), which enables the high-order mental processes (e.g., infer meaning) necessary for healthy cognition and behavior (Deco and Kringelbach, 2017; Park and Friston, 2013; Sporns, 2014; Zamora-López et al., 2010). Consistent with these theories, we found that a diminished capacity of
the brain to integrate information dynamically over time was associated with individuals who self-reported more frequent depressive symptoms (Chapter 2). The methods we applied rely on non-static representations of brain activity and thereby allow us to show that imbalances in whole-brain communication are accompanied by a more ‘rigid’ functional organization of the brain over both space and time. Our findings go beyond a growing body of literature on the role of temporal fluctuations in clinical populations (Kaiser et al., 2016) to and demonstrate that altered brain dynamics can already be found in association with depressive symptoms in nonclinical populations.

In contrast to measures of static functional connectivity, dynamic functional connectivity enables the identification of recurring patterns of connectivity by allowing brain regions to be temporally aligned. We explored how exactly the temporal features of functional connectivity can provide a better understanding of the complex brain dynamics underlying the mental processes. In Chapter 3, depression-related changes in functional connectivity were examined from both static and dynamic perspectives, using measures of time-averaged and time-varying functional connectivity, respectively. Both analyses embedded the role of the precuneus in depressive symptoms, consistent with other studies reporting aberrant functional connectivity of the precuneus in depressive patients. However, we observed that this extensive region is difficult to accurately study from a static functional connectivity perspective. This difficulty emerges from the multifaceted contributions of the precuneus to several other networks due to its involvement in a variety of cognitive processes including, but not limited to, visuospatial processing, episodic memory, and self-referential processes. We demonstrated that only the inclusion of the temporal dynamics of functional connectivity facilitates the disentanglement of the distinct configurations in which the precuneus plays an active part. Some of these connections might be able to help uncover the maladaptive cognitive processes associated with depressive symptoms.

Indeed, time-varying measures of functional connectivity have been recently shown to be more sensitive than static measures in capturing individual aspects of behavior (Eichenbaum et al., 2021; Rashid et al., 2014; Vidaurre et al., 2021). However, most neuroimaging studies on depression have investigated functional connectivity by assuming that it is static over the timescale of the recording session. This assumption may have limited our knowledge of the individual factors associated with the current state of the disease (Suo et al., 2018). In this thesis, the examination
of the temporal features in resting-state functional connectivity enabled us to successfully identify mechanisms that are specifically associated with the recurrence of a depressive episode (Chapter 4). By applying a method designed to track the expression of transient synchronization of distinct functional networks (Cabral et al., 2017a), we revealed that during the recurrence of a depressive episode, in comparison to the remission state, patients tend to increasingly engage in a brain state characteristic for the dysregulation of the reward network.

In summary, the findings of this thesis provide new evidence on the temporal evolution of functional connectivity, its importance for brain function, and its implications for depressive symptomatology in clinical and nonclinical populations.

### 6.3 Thesis limitations and further research directions

The initial set of goals of this research project were achieved, nevertheless, important considerations remain that require further discussion and follow-up investigations to facilitate a more in-depth understanding in future research.

The central goal of this thesis was to better understand the relationship between depressive symptoms and changing patterns of whole-brain connectivity, with special emphasis on the impact of depressive symptoms in the general population (individuals without a clinical diagnosis). We showed that individual mood responses after a relationship breakup represent an effective strategy to ensure variability in the levels of depressive symptoms among otherwise healthy individuals. This strategy was motivated by the large amount of evidence indicating that people after a stressful life event, often develop depression-like symptoms. Fortunately, individuals engage in cognitive and motivational strategies that allow them to maintain healthy levels of psychological functioning in the face of disruptive events. This capacity, known as resilience, is considered a good predictor of depression (Bonanno, 2004; Staudinger et al., 1995). However, a direct link between low levels of depressive symptoms after a breakup and the individual capacity to deal with negative events cannot be established in our studies. For example, individual differences in depressive symptoms may instead be the result of different levels of relationship engagement. It is also possible that some individuals did not perceive the termination of the relationship as a stressful event but as a pleasant relief. Therefore, categorizing a person who does not show symptoms of depression in the early months following the breakup as “resilient” seems inadequate.
Accordingly, an additional goal of this thesis was to investigate the specific brain adaptations that could help in the prediction of resilient responses to adverse circumstances. An experimental set-up was designed to capture over time the development of depressive symptoms after a relationship breakup. This study was interrupted due to COVID 19 regulations, leaving us with insufficient neuroimaging data to fully conduct the planned analysis. The results of the behavioral data are part of a publication (second author) and they are not included in this thesis. We found that individual mood-change trajectories can be categorized into four groups according to the shape of the trajectories: 1) 'resilient', with medium levels of depressive symptoms which decreased over time; 2) 'recovery', with high levels of symptoms that decreased gradually; 3) 'delayed', with moderate symptoms that increased over time; and 4) 'no recovery', with high levels of symptoms over the examination period. Continuation of this study can provide knowledge on how individual differences in resilience reflect depression-related imbalances in brain communication. We suggest that an understanding of how the brain adapts as a direct response to disturbing life events may assist in the development of novel strategies for strengthening resilience, thereby reducing the risk of developing depression.

A concurrent goal of this thesis was to elucidate the neuronal mechanisms associated with the occurrence of a new depressive episode when compared to the very same patients during a period of remission symptomatology (Chapter 4). The dynamical patterns of network connectivity were characterized using the same analytical approach as in Chapter 3, but here, nonclinical depression was investigated instead. This common approach across the two studies invites to further analyze whether the observed brain alterations associated with depressive symptoms in healthy adults could also explain the risk of relapse in currently asymptomatic patients with depression. Our results suggest that the neuronal factors associated with the development of depressive symptomatology in nonclinical populations may differ from those in currently diagnosed patients. However, as the repertoire of brain states was defined in a data-driven way, resulting in a unique set of brain states for each study, a direct comparison between the two populations was hindered. We propose that subsequent investigations should consider a repeated two-by-two study design, where two groups, a clinical and a nonclinical, are measured twice, with and without present symptoms, to ensure comparability. This setup will deepen the understanding of the
interrelationship between depressive symptoms and clinical diagnosis. It is important to note that depressive symptoms are not unique to MDD. Not only can they be seen in healthy individuals but across a wide range of psychiatric and neurological illnesses such as anxiety, dementia, Parkinson’s disease, and schizophrenia. This calls for a cross-diagnostic research approach that investigates how the neuronal mechanisms that underlie depressive symptoms overlap across these illnesses.

The impact of depressive symptoms (in both clinical and nonclinical populations) on the patterns of brain functional connectivity was investigated by scanning individuals in a resting-state. Even in the absence of an externally imposed task, such as in rest, individuals engage in a wide range of complex patterns of self-generated thinking (Berman et al., 2011; van der Meer et al., 2010), and these widely heterogeneous thinking patterns constitute an essential feature of human cognition (Christoff et al., 2016). We specifically chose the resting-state over the task state to capture the inner experience while individuals freely engage in several forms of mental activity — unconstrained by the explicit demands of the task. Furthermore, studying the brain at rest may provide a richer characterization of brain activity than during task performance (Ponce-Alvarez et al., 2015). Despite these advantages, one of the pitfalls is the absence of experimental control. The only instruction that the participants received was to lie still and let the mind flow. Raising the critical question of what do subjects do when they ‘do nothing’? Previous literature has shown that individuals with depressive symptoms have more difficulties avoiding rumination when they are free to think. This tendency of rumination has been associated with abnormal interactions within the default mode network. Moreover, between this network and the rest of the brain. However, rumination is often measured as a trait-construct, that is as a habitual reaction to a sad mood. This leaves the possibility that high trait ruminators might not necessarily be ruminating during the resting-state session. Therefore, it seems necessary to account for individual differences regarding the experience of the resting-state recording session. These differences may be related to specific thinking patterns such as rumination, but also to individual differences regarding difficulties of being motionless, resisting achy sensations, fighting fatigue, or focusing on background noises. A possible way to address this issue is the recording of a questionnaire related to the experiences during the scan at the end of the session, while the individual is still inside the scanner.
Finally, although the advances in neuroimaging methods and functional connectivity analysis provide powerful tools to the investigation of the neuronal basis of depression, the results across studies vary significantly (Gray et al., 2020; Müller et al., 2017; Suo et al., 2018). With the rapid growth in the quantity of recorded neuroimaging data and the variety of analytical developments to investigate fMRI brain activity, it is becoming increasingly difficult to compare results across studies. Moreover, this problem may be in part due to the differences concerning fMRI acquisition parameters, processing procedures (i.e., temporal filtering, global signal regression), brain parcellation strategies, and the utilized analytical tools in brain connectivity estimation. On the other hand, the heterogeneous findings may also be attributed to the multifaceted nature of the disorder that encompasses diverse disease subtypes, symptomatology, and treatment responsiveness. Nevertheless, the sometimes-weak correspondence between the diagnosis and neuronal substrates raises questions about the adequacy of the current approach in diagnosing mental disorders, namely the Diagnostic and Statistical Manual of Mental Disorders (DSM) and International Classification of Diseases (ICD). Although knowledge is needed toward the identification of the precise neuronal markers for the assessment of the risk of depression, neuroimaging research provides a valuable tool to challenge current conceptualizations of MDD and assists in the search for targeted treatments.

6.4 Conclusion

Owing to the rapid growth of the field of neuroscience, important progress has been made in elucidating the neuronal mechanisms involved in MDD. However, we still do not fully understand how these mechanisms originate in the healthy brain and how they contribute to future relapses. This thesis suggests that the temporal properties of whole-brain functional connectivity provides additional information that may be meaningful to understand the relationship between brain dynamics and depression symptoms. We showed that irrespective of the clinical diagnosis, the present experience of depressive symptoms was directly linked to imbalances in the dynamic orchestration of interacting brain networks. Therefore, we underscore the importance of investigating the neural mechanisms associated with depressive symptoms in the general population as it provides new opportunities for identifying depression vulnerability and, ultimately, anticipating the risk of MDD.