From micro to macro

Impact Paragraph

"If all you have is a hammer, everything looks like a nail."
-Abraham Maslow, "The Psychology of Science", 1966

This thesis describes a series of experiments which advance our understanding of a widely used neuroscientific and therapeutic tool, Transcranial Magnetic Stimulation (TMS). In addition to the scientific and societal impact of the results described, this thesis is an example of approaching a research question from different perspectives and combining different disciplines, expertise, and research techniques to approach scientific bottlenecks. It is the first funded project in a newly established collaboration between the Faculties of Psychology and Neuroscience (FPN) and Health Medicine and Life Sciences (FHML) called the "Centre for Integrative Neuroscience (CIN)". This initiative has allowed for the bridging of research, expertise, techniques and communication between two different neuroscience disciplines at Maastricht University.

The research and main findings described in this thesis are important for advancing our understanding of how TMS exerts its effects. TMS is a form of non-invasive brain stimulation, which is widely used in neuroscience research around the world. It uses electromagnetic pulses to briefly and painlessly send electricity into the brain (1). For example, when a pulse is given over the motor cortex, it can directly activate the neurons beneath, carrying an electrical signal along a specific neural circuit to a finger muscle, and causing a visible finger twitch (2). When many of these pulses are repeated in a specific pattern (as repetitive, rTMS), longer-lasting stimulation effects can be induced, including increased or decreased activity of a particular brain region (3). In other words, giving a short round of rTMS over a particular brain region can deactivate or increase activity in that specific brain region for a short time after the stimulation is over. This is very useful in understanding what role a particular brain region plays in certain processes.

Perhaps the most important use of rTMS is in the clinic, where it is a treatment option for several psychiatric and neurological disorders. It is most commonly used as a treatment for depression (4-6), but obsessive-compulsive disorder (OCD), pain and stroke are some other examples of disorders being treated with rTMS (7). Despite the widespread use of rTMS both in research and therapeutically, the underlying mechanisms are relatively unknown. Understanding the very basic, molecular machinery which underly rTMS effects would allow us to design stimulation patterns to be the most safe and effective for research and clinical treatment. With advances in neuroscientific methods, it may even be possible personalize treatment protocols, but only with a strong background of research identifying reliable molecular targets of rTMS effects.

This thesis takes a unique, inter- and cross disciplinary approach to understanding the underlying molecular mechanisms of TMS. It is interdisciplinary for combining studies on a molecular level using human neurons grown in the lab (**Chapters 2-4**), with rTMS studies in human participants (**Chapters 6-8**). Several important findings are reported in these chapters, and they contribute to a better understanding of how rTMS is able to create lasting effects in humans. We show in **Chapter 3** that human neurons stimulated with different rTMS protocols respond in the expected way. Neurons stimulated with an rTMS protocol thought to *increase* the activity of a particular brain region were more strongly activated than neurons stimulated with an rTMS protocol thought to *decrease* it. Similarly, in **Chapter 4** we show evidence for increased expression of a few important genes after stimulation with the excitatory protocol, but more research is needed on the specific molecular pathways activated by the different stimulation protocols. In humans, we show that it can be difficult to replicate the expected effects of stimulation due to many sources of variability, which again supports that future research in human cellular models such as in Chapters 3 and 4 is important. We hypothesize that combining multiple stimulation sessions could enhance stimulation effects, and in the final chapter (**Chapter 8**) provide preliminary evidence that stimulation at one area of the brain could lead to activation of different, remote brain areas. Overall, the findings of this thesis both in human neuronal cell culture and in human participants add to our understanding of the underlying mechanisms of TMS, and offer suggestions for future research in this area.

One of the larger, societal impacts of these findings lies in the therapeutic potential of TMS. We are currently in the middle of a serious global pandemic, where many are forced to self-isolate, work from home and are burdened with financial and health worries. This understandably has a large impact on the mental health of millions worldwide, with likely consequences such as an increase in the global burden of depressive disorders for years to come (8).

Depression is one of the most severe mental health disorders, ranked by the WHO as the single largest contributor to global disability (9), and with several large studies consistently placing it within the highest for disease burden and disability adjusted life years (DALYS) (10-12). Depression therefore has a huge impact on the quality of lives of millions of people worldwide, having substantial social and economic consequences. In addition to severely reducing the quality of lives of people suffering from depression, the economic costs of depression are huge. Global estimates of costs due to lost productivity are in the billions (US dollars) (13), not to mention the burden on the healthcare system. This indicates the critical need for an effective and quick treatment option. The findings of this thesis highlight the potential of a short-duration rTMS protocol: intermittent Theta Burst Stimulation (iTBS), which requires only 3 minutes to apply, and has also been shown to be effective as a treatment for depression (14). iTBS is additionally promising as it is quick, and therefore could be

used to treat many people per day. This thesis provides support for iTBS as a treatment protocol, in particular when repeated several times in a single day (as accelerated iTBS). A review on this idea is also included as a chapter in this thesis.

There are several steps required for the findings described in this thesis to have a clinical impact, not only in the treatment of depression but also in the treatment of other mental and neurological disorders. First, more research needs to be done on the efficacy of accelerated iTBS with longer intervals (50-60 minutes). For instance, research with healthy participants in a similar setup as described in **Chapter 6**, but as proposed in **Chapter 5**, with three iTBS sessions separated by 50-60 minutes. If this is proven effective in healthy participants, large clinical trials could test whether it is also as effective as a treatment. Some potential disorders where this could be effective could be in cognitive decline and dementia, where patients could do cognitive enhancement tasks (memory games, reading or drawing tasks) during the long breaks between iTBS sessions. Similarly, as a treatment for depression, Cognitive Behavioral Therapy (CBT) could be done in the breaks between iTBS sessions, as this combination has proven to improve treatment efficacy (15).

At the molecular level, further research following up on the findings of this thesis using TMS and human neuronal cells is needed. Identifying specific, molecular mechanisms of rTMS effects could help us to understand why some people respond well to rTMS treatment, but others do not. This thesis provides evidence for calcium imaging being a potential indicator of iTBS efficacy (**Chapter 3**), but further research would be needed to confirm this. Studies could use patient-derived neurons to indicate whether a particular patient would be responsive to a particular stimulation protocol. For example, if a particular cellular response (such as calcium imaging) can indicate responsiveness to a certain rTMS protocol, then patients could have their neurons tested for responsiveness before undergoing rTMS treatment. Much more research is needed for this to be a realistic option in the future; however, this thesis provides a step in this direction towards identifying molecular targets to indicate rTMS efficacy, and to use these targets as predictors for patient responsiveness to treatment.

Future studies can use more advanced human neuron models to investigate different molecular targets, and can work with other TMS users from different disciplines to better understand the limitations and main research questions in their research areas. Combining input from experts in computational modelling, engineers and physicists who can work to develop optimal TMS coil designs, and researchers/clinicians who use TMS on human participants or can establish a neuronal cell model are all important in advancing our understanding of how TMS works. In conclusion, this thesis provides an example of the benefits of interdisciplinary research, and describes several important findings which have a larger societal and clinical impact. Notably in

the treatment of depression, but also for other applications, where support for accelerated treatment protocols as well as molecular targets for assessing stimulation responsiveness are reported. Future interdisciplinary research into these ideas will lead us to gain deeper insights into the underlying mechanisms of rTMS, and optimize its use in research and the clinic.

References

1. Barker AT, Jalinous R, Freeston IL. Non-invasive magnetic stimulation of human motor cortex. Lancet. 1985;1(8437):1106-7.
2. Rothwell JC, Hallett M, Berardelli A, Eisen A, Rossini P, Paulus W. Magnetic stimulation: motor evoked potentials. The International Federation of Clinical Neurophysiology. Electroencephalogr Clin Neurophysiol Suppl. 1999;52:97-103.
3. Hallett M. Transcranial magnetic stimulation: a primer. Neuron. 2007;55(2):187-99.
4. George MS, Wassermann EM, Williams WA, Callahan A, Ketter TA, Basser P, et al. Daily repetitive transcranial magnetic stimulation (rTMS) improves mood in depression. Neuroreport. 1995;6(14):1853-6.
5. O'Reardon JP, Solvason HB, Janicak PG, Sampson S, Isenberg KE, Nahas Z, et al. Efficacy and safety of transcranial magnetic stimulation in the acute treatment of major depression: a multisite randomized controlled trial. Biol Psychiatry. 2007;62(11):1208-16.
6. McClintock SM, Reti IM, Carpenter LL, McDonald WM, Dubin M, Taylor SF, et al. Consensus Recommendations for the Clinical Application of Repetitive Transcranial Magnetic Stimulation (rTMS) in the Treatment of Depression. J Clin Psychiatry. 2018;79(1).
7. Lefaucheur J-P, Aleman A, Baeken C, Benninger DH, Brunelin J, Di Lazzaro V, et al. Evidence-based guidelines on the therapeutic use of repetitive transcranial magnetic stimulation (rTMS): An update (2014–2018). Clinical Neurophysiology. 2020;131(2):474-528.
8. Xiong J, Lipsitz O, Nasri F, Lui LMW, Gill H, Phan L, et al. Impact of COVID-19 pandemic on mental health in the general population: A systematic review. J Affect Disord. 2020;277(1573-2517 (Electronic)):55-64.
9. Organization WH. Depression and Other Common Mental Disorders: Global Health Estimates. Geneva: World Health Organization; 2017. Contract No.: Licence: CC BY-NC-SA 3.0 IGO.
10. Murray CJL, Lopez AD. Alternative projections of mortality and disability by cause 1990–2020: Global Burden of Disease Study. The Lancet. 1997;349(9064):1498-504.
11. Ferrari AJ, Charlson FJ, Norman RE, Patten SB, Freedman G, Murray CJL, et al. Burden of Depressive Disorders by Country, Sex, Age, and Year: Findings from the Global Burden of Disease Study 2010. PLOS Medicine. 2013;10(11):e1001547.
12. James SL, Abate D, Abate KH, Abay SM, Abbafati C, Abbasi N, et al. Global, regional, and national incidence, prevalence, and years lived with disability for 354 diseases and injuries for 195 countries and territories, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. The Lancet. 2018;392(10159):1789-858.
13. Chisholm D, Sweeny K, Sheehan P, Rasmussen B, Smit F, Cuijpers P, et al. Scaling-up treatment of depression and anxiety: a global return on investment analysis. The Lancet Psychiatry. 2016;3(5):415-24.
14. Blumberger DM, Vila-Rodriguez F, Thorpe KE, Feffer K, Noda Y, Giacobbe P, et al. Effectiveness of theta burst versus high-frequency repetitive transcranial magnetic stimulation in patients with depression (THREE-D): a randomised non-inferiority trial. Lancet. 2018;391(10131):1683-92.
15. Donse L, Padberg F, Sack AT, Rush AJ, Arns M. Simultaneous rTMS and psychotherapy in major depressive disorder: Clinical outcomes and predictors from a large naturalistic study. Brain Stimulation: Basic, Translational, and Clinical Research in Neuromodulation.