

VU Research Portal

Framing depression in a SN(a)Pshot

Woudstra, S.

2013

document version

Publisher's PDF, also known as Version of record

[Link to publication in VU Research Portal](#)

citation for published version (APA)

Woudstra, S. (2013). *Framing depression in a SN(a)Pshot: Imaging risk factors of MDD.* [, Vrije Universiteit Amsterdam].

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal

Take down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

E-mail address:

vuresearchportal.ub@vu.nl

Chapter 6

General discussion

Partly based on:

Stein JL, Medland SE, Vasquez AA, Hibar DP, Senstad RE, (...), **Woudstra S**, et al. (2012) *Identification of common variants associated with human hippocampal and intracranial volumes. Nature Genetics 44(5):552-561*

Bis JC, DeCarli C, Smith AV, van der Lijn F, Crivello F, (...), **Woudstra S**, et al. (2012) *Common variants at 12q14 and 12q24 are associated with hippocampal volume. Nature Genetics 44(5):545-551*

Understanding the mechanisms underlying major depressive disorder (MDD) is key for developing personalized treatment programs. Getting closer to this understanding can amongst others be established by identifying genetic risk factors that are associated with intermediate phenotypes of (subtypes of) MDD (e.g. PCLO genotype with altered emotion processing). The overall aim of the studies presented in this thesis was to identify imaging characteristics associated with the occurrence and recurrence of depression. This was investigated from two perspectives: Using a neuroimaging genetics approach in this thesis we identified the PCLO risk allele to modulate amygdala function during fearful facial processing in MDD, which may increase the vulnerability for the development and prolonged course of MDD. In addition, susceptibility for depression may also be increased by the PCLO risk allele by modulating local brain in responses to salient stimuli (i.e. insula) and processing novel negative information. Using a longitudinal design in this thesis we identified subjects at risk for non-remitted outcome using regional volume and EF abnormalities as predictors (i.e. smaller inferior frontal, and cingulate volumes as well as increased dlPFC activity).

Summary

MDD is characterized by affective symptoms and cognitive impairments, and has been associated with biased memory formation for mood-congruent information, which in turn may reflect changes in limbic and prefrontal activity as well as altered monoaminergic neurotransmission. The piccolo (PCLO) gene, involved in monoaminergic neurotransmission, has previously been linked to depression in a genome-wide association study. In **chapter 2** we showed that the PCLO risk allele is associated with increased activity in the left amygdala during processing of negative faces, but not positive faces. No association with executive function or its neural substrate was found. The paradigms used in this chapter are considered to represent 'pure' examples of tests probing emotion processing and executive function, respectively. Hence, PCLO is probably mainly involved in primary emotion processing, and not in tasks that recruit brain areas involved in executive functioning, emotion regulation, or in implementing action programs important for motivational problems of depression. This indicates that PCLO may act as a risk factor for MDD predominantly through stronger amygdala reactivity and, possibly, altered serotonergic neurotransmission in brain areas related to emotional information processing. A combined cognitive-emotional paradigm was employed in **chapter 3** to further investigate whether the PCLO risk allele was associated with the neural substrate of memory processing of positive and negative information. During negative word encoding, risk allele carriers showed significantly lower activity relative to non-risk allele carriers in the insula, and trend-wise in the anterior cingulate cortex and inferior frontal gyrus. Moreover, depressed risk allele carriers showed significantly lower activity relative to non-risk allele carriers in the striatum during negative word encoding, an effect which was absent in healthy controls. Finally, amygdala responsiveness during processing new positive words vs. known words was blunted in healthy PCLO+ participants and in MDD patients irrespective of genotype, which may indicate that signalling of salient novel information does not occur to the same extent in PCLO+ subjects and MDD patients. The PCLO risk allele may increase vulnerability for MDD by modulating local brain function with regard to responsiveness to salient stimuli (i.e. insula) and processing novel negative

information. Also, depression-specific effects of PCLO on dorsal striatal activation during negative word encoding and the relative absence of amygdala salience signalling for novel positive information further suggest a role of PCLO in symptom maintenance in MDD. Summarizing, the results obtained in part I emphasize the modulating role of PCLO during emotion processing in MDD. Moreover, PCLO may modulate the responsiveness to salient stimuli in an emotional context, whereas it does not influence executive dysfunction in MDD.

The second part of this thesis aimed to provide insight into the course of neuroanatomical and functional brain abnormalities in MDD in a longitudinal study. Recurrence of depressive episodes in MDD is a burden for both patients and society and executive impairments may increase the risk for recurrent depression. Early prediction of recurrence is therefore key for prevention. Non-remitted MDD patients showed smaller right inferior frontal, rostral anterior cingulate, and posterior cingulate volumes than healthy controls at both baseline and after two-year follow-up. These smaller volumes were not observed in remitted patients (**chapter 4**). Investigation of the role of executive function (EF) abnormalities in predicting unfavourable course (**chapter 5**) revealed increased right dorsolateral prefrontal (dlPFC) activity as predictor for non-remitted outcome. No evidence for aggravation of EF abnormalities associated with unfavourable course was found. Taken together, volume abnormalities could potentially serve as biological markers for poor clinical outcome, and increased right dlPFC activity may distinguish those at risk for non-remission from those with a more favourable course. These results indicate a possible step in MRI-based complementary MDD staging but future long term follow-up functional neuroimaging treatment studies are necessary to investigate the role of increased dlPFC activity on treatment response prediction for improvement of individual-based treatment programs.

Methodological considerations

Part I

PCLO may be considered a new candidate gene for depression (or more generally disturbed affective processing) as it appears to modulate local brain function during emotion processing and emotional memory processing, but not during executive function. This conclusion implies that pathways to MDD have been further elucidated by the results from chapter 2 and 3. However, some limitations have to be considered.

First, can it be assumed that only one SNP of the entire PCLO gene affects the function of this large protein? The results of several studies, further investigating the role of this SNP (rs2522833) in MDD, may provide an answer to this question. Furukawa-Hibi and colleagues found evidence for the SNP to affect the protein up to the behavioural level: they created transgenic mice over-expressing the C2A domain of PCLO – the domain containing the non-synonymous coding SNP rs2522833 –, showing depression-like behaviour in several tests, indicating that PCLO indeed may modulate depressive behaviour on its own (Furukawa-Hibi *et al.*, 2010). Schuhmacher and Kuehner (Kuehner *et al.*, 2011; Schuhmacher *et al.*, 2011) provided evidence for the SNP to modulate the hypothalamic–pituitary–adrenal (HPA) regulation, which is known to be involved in stress and has often been associated with depression (Knorr *et al.*, 2010; Penninx *et al.*, 2013; Vreeburg *et al.*, 2009). Schuhmacher *et al.* found that risk allele carriers showed increased HPA dysregulation after antidepressant treatment. These

results suggest that an affected PCLO protein may enhance the biological response to SSRIs among MDD patients. Kuehner also found a dysregulated HPA system in PCLO risk allele carriers, measured using the cortisol awakening response (CAR; Kuehner *et al.*, 2011). This finding indicates that an affected PCLO protein may exhaust regulatory mechanisms of the HPA system. The PCLO genotype has also been associated with personality traits, including neuroticism (Kuehner *et al.*, 2011) and harm avoidance (Minelli *et al.*, 2012). These traits have been shown to increase the risk for MDD (Farmer, 2003; Klein *et al.*, 2011). Another SNP in the piccolo gene has additionally been associated with bipolar disorder, further suggesting that PCLO plays an important role in mood disorders (Choi *et al.*, 2011). Together with our results, these findings indicate that the PCLO risk allele likely affects the protein structure of PCLO, resulting in a cascade of alterations related to pathophysiological mechanisms underlying MDD, such as changes in local brain activity.

Second, although this PCLO risk allele (rs2522833) was identified in a GWAS for MDD with eleven top-200 signals localized to a 167 kb region overlapping the PCLO gene, it did not reach significance levels (Sullivan *et al.*, 2009). Further investigations showed that among these association signals within the PCLO gene, the rs2522833 polymorphism, located near a C2 calcium-binding domain of the PCLO protein, was most closely associated to MDD (Bochdanovits *et al.*, 2009). Findings from other GWA studies were inconsistent: some could replicate the PCLO findings, whereas others did not (Hek *et al.*, 2010; Shyn *et al.*, 2011; Wray *et al.*, 2012). Also, the psychiatric genome consortium (PGC), using >9000 cases and >9000 controls could not replicate the PCLO finding (Major Depressive Disorder Working Group of the Psychiatric GWAS Consortium, 2013). These results also reflect the high heterogeneity and moderate heritability of MDD. Moreover, the lack of significance in the GWAS further highlighted the need for a phenotype that is closer to the biological substrate of MDD than the clinical diagnosis itself. Despite the fact that no SNP in the PCLO gene was significantly associated with MDD, researchers showed PCLO to be a modulating factor in depression (Furukawa-Hibi *et al.*, 2010; Kuehner *et al.*, 2011; Minelli *et al.*, 2012; Schuhmacher *et al.*, 2011). These results also imply that the follow-up of GWAS data using neuroimaging data as intermediate phenotype, may lead to the identification of new promising candidate genes for MDD.

Part II

Early identification of subjects at risk for the recurrence of depression is essential, since circa 50% of MDD patients experience recurrence of MDD within five years after an episode, and 80% of patients with a history of two episodes will have another recurrent depression (Burcusa & Iacono, 2007). Early prediction of recurrent depression is key for selecting patients for relapse preventing therapy. However, the pathogenetic mechanisms underlying recurrence are unclear, and useful biomarkers are lacking. Neuroimaging abnormalities are considered to be a stable neuropsychological trait characteristic, present in both chronic depression and remission. However, neuroimaging studies (e.g. Lorenzetti *et al.*, 2009; Maalouf *et al.*, 2011) were often performed cross-sectionally, and were, therefore, unable to infer on effects of allostatic disease-load, or to disentangle state and trait effects of the disease.

An intermediate phenotype can be considered a *state* characteristic of MDD when it is present during the depressive episodes only, and is absent in remission (van Wingen *et al.*, 2010b). The main feature of a *trait* characteristic is that it is present independent of current diagnostic status (e.g. remission or recurrence). Thus, trait characteristics of depression may already be present before the onset of the disorder. Using a longitudinal neuroimaging design we found our intermediate phenotypes not to be trait characteristics of depression, but rather to be specific for those patients at risk for an unfavourable course. This indicates that the disease profile characteristics should be accounted for when searching for intermediate phenotypes, such as smaller regional brain volumes. To date, the studies described in part II of this thesis are the first whole-brain longitudinal structural and functional MRI studies in MDD to show that IFG, rostral ACC, and PCC volumes as well as increased dlPFC activity are predictive of non-remitted course, and these results may be used for selecting patients for relapse preventing therapy.

No longitudinal differences

Based on the cross-sectional results of our data (van Tol *et al.*, 2011; van Tol *et al.*, 2010), we expected to find longitudinal changes in volume (**chapter 4**) and aggravation of EF abnormalities (**chapter 5**). However, these hypotheses were not confirmed by our results. Previous longitudinal studies are in line with our findings (i.e. no progressive reduction) (Frodl *et al.*, 2008a; Soriano-Mas *et al.*, 2011), although results have been inconclusive (even within research groups) (Frodl *et al.*, 2008c). Two possible explanations may account for the lack of longitudinal effects. On the one hand, we expected progressive atrophy in recurrent depression relative to remitted depression, as reflected in longitudinal changes. We also expected normalization of EF abnormalities at S2 in remitted patients, together with aggravation of EF abnormalities at S2 in non-remitted patients. Instead, we found smaller volumes and EF abnormalities in non-remitted patients at S1, implying that these may serve as predictors for non-remitted outcome. On the other hand, longitudinal changes could have been present, but due to the time period of two years, we might not have been able to detect these. Also, disease load of the participants may have been of influence. As participants were mildly to moderate depressed, it may take a longer time period to detect changes at the regional brain volume and neuronal activity level. The trait like abnormalities found in (van Tol *et al.*, 2011; van Tol *et al.*, 2010) therefore likely reflect a vulnerability factor in patients who recovered from a depressive episode. Follow-up of the patients and healthy controls over a longer time period (e.g. > 2 years) may provide an answer to the question whether longitudinal changes occur over a longer time period.

Another possible limitation of the longitudinal neuroimaging studies is that no neuroimaging data of course trajectories were available. When available, these data would help identifying the course direction of the effects, and would elucidate whether the predictive values we found would be gradually formed over two years.

General considerations

Our studies were embedded in the Netherlands Study of Depression and Anxiety (NESDA study), which aims to investigate the naturalistic course of depression. NESDA has a multi-

site naturalistic cohort design with a longitudinal framework. It further aims to integrate biological and psychosocial research paradigms within an epidemiological approach in order to examine the predictors of the long-term course and consequences (Penninx *et al.*, 2008). Being embedded in the NESDA study allowed us to include a large number of subjects, taking into account a variety of potentially confounding factors. However, some issues should be considered.

Depression severity in the MDD patients in the NESDA MRI study was only mild to moderate, due to recruitment from the general population, general practitioners, and outpatient mental health organizations, but not from inpatient clinics. Consequently, we do not know whether our interaction findings would have been even more robust when inpatients had also been included.

The neuroimaging data was gathered from three different sites, based in Amsterdam (AMC), Leiden (LUMC), and Groningen (UMCG). We used similar 3T systems at each site in this multicenter study, and no systematic scanning site x diagnosis bias was observed. However, variability in image acquisition may have occurred due to minor differences in hardware (receiver coil), imaging parameters, and timing of software upgrades.

Although we investigated the effect of SSRI treatment by repeating the analyses in a subset of medication free participants, due to the naturalistic design, we were not able to investigate the effects of SSRI treatment in an experimental setting, such as an intervention study.

Does the scar hypothesis still stand

One theory on the association between MDD and structural changes is that smaller volume of limbic areas, such as amygdala, insula, hippocampus (Frodl *et al.*, 2008c; Hatton *et al.*, 2012; MacQueen *et al.*, 2003) and prefrontal regions, such as ACC and dorsal medial PFC (Frodl *et al.*, 2008c), may result from prolonged duration of the disorder (i.e., ‘scarring’ (McEwen, 2001; Sheline, 2000)), due to chronically elevated levels of stress hormones (Post *et al.*, 2012) and possibly abnormal BDNF levels (Gonul *et al.*, 2010). Such scarring could explain the heightened vulnerability for new depressive episodes, as these morphological abnormalities may underlie functional impairments in emotion processing and mood regulation. In our studies, we found no evidence for the scar hypothesis, i.e. absence of progressive decline over two years. However, smaller volumes may still arise from a more prolonged duration of the disorder, or could occur up to a certain level and then halt. In addition, scarring may be detected only in patients with severe depression, which were not present in our sample. Long-term prospective studies are needed to fully elucidate these issues.

Alternative hypothesis

An alternative hypothesis to the scar hypothesis would be that structural abnormalities and EF impairments in fact represent a premorbid vulnerability factor, presumably associated with genetic or familiar risk for depression, that can be regarded a predictor rather than a consequence of a chronic course. We found some evidence that smaller IFG and cingulate volumes as well as increased dlPFC activity, can indeed be regarded as predictors for non-remitted and unfavourable outcome. To fully corroborate the predictive value of these

abnormalities, an extra group should be added to the longitudinal study design. This group should consist of participants who develop depression within the study. Unfortunately, the number of these participants in our study was too low ($n=2$).

The main aim of this thesis was to identify imaging characteristics associated with the occurrence and recurrence of depression. An additional aim was to enhance understanding of the underlying neurobiological mechanisms of MDD. Although the character of this thesis is fundamental by origin, some findings may have clinical implications.

Clinical implications

Antidepressant medication is widely available and is used by circa one million people in the Netherlands (CBS, 2011). However, only 60% of treated patients show sufficient response to medication and side effects are common. In clinical practice, several options are available for treating a patient's insufficient response to an initial antidepressant dose. These options include increasing the dose, switching to another antidepressant, or combining several drugs (Berney, 2005). Moreover, as antidepressant pharmacotherapies are based on outcome averages from large clinical trials, patients would likely benefit from treatment based on personal medication tolerance and response profiles and specific course predictors.

A more personalized approach, based on specific genetic information of the patient, could increase treatment success. The sequencing of the human genome in the beginning of this millennium (Venter *et al.*, 2001) was expected to revolutionize medicine and to result in individualized treatments based on the genetic make-up of the patient. To date, genetic studies of MDD have not yet led to diagnostic and treatment biomarkers (Miller & O'Callaghan, 2013). However, there is progress in determining the role of the genome in drug metabolism, which is the first effort in personalized antidepressant medication. The knowledge on the role of the PCLO risk allele, modulating emotion processing and memory of negative information, which is associated with increased amygdala and insula activity, may be used in developing personalized antidepressants based on the PCLO genotype. Moreover, the PCLO genotype in combination with intermediate phenotypes may potentially serve as biomarker that eventually leads to better relapse prevention.

Clinical implications for neuroimaging results seem to be more straightforward than for neuroimaging genetic results. The neuroimaging results described in this thesis may eventually be used on an individual level to predict prognosis and may be used for targeting brain areas with treatment. In the following paragraphs these two options are discussed.

Use of neuroimaging parameters as predictor for prognosis

We found smaller volumes of inferior frontal and cingulate volumes to be predictive of poor depression outcome. In theory the information of a single patient could be used for personalization of treatment. Single-subject MRI scans to measure the volume and morphology of the brain together with standard clinical tests to diagnose MDD (e.g. CIDI) would, theoretically, provide the information for a practitioner needed to personalize the treatment. However, one should realize that results are based on group level statistics, and variation among individuals is large. In order to implement the predictive results described in this thesis on a single subject level, future studies should focus on the sensitivity and specificity of structural MRI data for individual prediction. This is one of the key-points NESDA is currently working on.

Use of neuroimaging for neurobehavioral therapy

Functional MRI results, such as our increased right dlPFC activity during planning, may be suitable as a treatment target. The term neurobehavioral therapy is used to describe treatment that directly acts on the underlying mechanisms (i.e. activation or inhibition of depression related brain areas; Siegle *et al.*, 2007). Because this type of therapy is specifically designed with the underlying (biological) mechanisms in mind, the expectation is that it may eventually lead to symptom reduction. Neurobehavioral therapy may especially be useful for treatment resistant depression or for those with an unfavourable outcome prognosis; conventional therapies like psychological or pharmacological therapy have a high drop-out ratio in the treatment resistant group (>25%) and often recovery is incomplete (Siegle *et al.*, 2007), indicating that more specific treatment are necessary to obtain higher recovery rates. Neurofeedback using fMRI data is an example of neurobehavioral therapy and can be used by patients to directly modify neural function by monitoring personal brain function markers (i.e. self-control) with the goal of changing them (Keedwell & Linden, 2013). The first clinical pilot study using neurofeedback in depression tested the hypothesis whether self-control of positive emotion areas would promote the experience of such emotions and enhance patients' experience of self-efficacy (Linden *et al.*, 2012). The results were promising, and our results may initiate an investigation to answer the question whether neurofeedback training targeting dlPFC regulation during executive function may contribute to a better clinical outcome. The optimal neurobiological treatment outcome here would be lowering the increased right dlPFC activity in non-remitted patients together with improvement of planning and concentration. Although the pilot study of Linden and colleagues and the clinical implications of our results appear to be promising, formal clinical efficacy studies (i.e. randomised blinded clinical trials) are needed to exclude possible placebo effects and to judge the therapeutic potential of fMRI-based neurofeedback as useful adjunct to current therapies for depression. Furthermore, our findings may be used in transcranial magnetic stimulation (TMS) therapy for treatment of depression. Repetitive TMS (rTMS) treatment is a relatively new, but robust treatment method for MDD (Leuchter *et al.*, 2013) and is currently used only on patients resistant for antidepressant medication. rTMS enhances cortical excitability and has shown antidepressant effects when applied over the left PFC (Schutter & van Honk, 2005). The cortical excitability induced by rTMS may be used to lower the effects of increased right dlPFC, which we found to be predictive for poor clinical outcome, and therefore be used as preventive treatment.

Recommendations for future research

We have shown that the follow-up of GWAS data, using neuroimaging data as intermediate phenotype, may lead to the identification of new promising candidate genes for MDD, such as PCLO. To further investigate the role of PCLO in emotion processing, a pathway analysis approach is promising. As shown for schizophrenia, the identification of groups of genes with similar cellular function rather than single genes may have better clinical implications for finding additional drug targets (Lips *et al.*, 2012). Also the use of genome-wide association of neuroimaging data may be promising to confirm the role of PCLO in MDD. Recently, we

showed genome-wide association for hippocampal volume in a meta-analysis of GWA studies in healthy subjects (Stein *et al.*, 2012; Bis *et al.*, 2012). Future genome-wide association meta-analyses should be performed in MDD patients to strengthen the association of PCLO with MDD.

In order to develop personalized medicine based on PCLO genotype, future studies should focus on the role of PCLO in especially the serotonergic pathway. A promising approach is likely the use of positron emission tomography (PET) tracers to study radioligand binding to receptors that are influenced by the presynaptic PCLO protein, as has previously been shown in other studies on genes associated with serotonin transporter function (David *et al.*, 2005). In addition, the use of longitudinal MRI designs may be helpful to investigate whether healthy PCLO+ subjects that show a negative bias reflected in lower frontostriatal activity are indeed more vulnerable to develop MDD.

Clearly, in order to use the neuroimaging data from **chapter 4** and **5** for neurobehavioral therapy or as predictor for prognosis, future research should focus on the sensitivity and specificity of the MRI data for individual prediction. Moreover, replication of our findings is needed, preferably across in- and outpatients. In addition, future studies, using a longitudinal design that specifically investigates treatment response, should elucidate the predictive value of increased dlPFC activity. Finally, future research should investigate whether smaller volumes of brain areas may also predict a poor clinical outcome in high-risk groups, such as subthreshold MDD, and groups with a high familial load.