Serotonin, cortisol, and stress-related psychopathology

Tanke, Marit Aline Christine

Chapter 10

Summary & General discussion

SUMMARY

The aim of this thesis was to study the role of low serotonergic activity and a dysregulated hypothalamus-pituitary-adrenal axis (HPA-axis) in vulnerability to stress and stress-related psychopathology. In the animal studies, we experimentally induced changes in the brain serotonergic system or in the HPA-axis and evaluated physiological and behavioral stress parameters. The clinical studies focused on conditions likely to be associated with long-term alterations in serotonin and/or HPA axis function. This approach allowed investigation of the associations between serotonin function, HPA-axis function, stress susceptibility, and emerging psychiatric symptoms.

Part I: animal studies

Serotonergic system

Changes in serotonin (5-HT) function may lead to an altered stress response. To study this relation in more detail we have attempted to change serotonin function by dietary depletion of tryptophan, also in combination with specific 5-HT receptor agonists, and discontinuation of chronic Selective Serotonin Reuptake Inhibitor (SSRI) treatment.

Dietary tryptophan depletion reportedly decreases central 5-HT function (47). In **chapter 2,** animals on a tryptophan deficient diet displayed significantly higher corticosteroid levels than those on a normal diet. In addition, daily immobilization stress increased the adrenal weights and the reactivity to acoustic stimuli. Habituation to the acoustic stimuli was not affected by tryptophan depletion. This indicates that although a low-tryptophan diet increases the sensitivity of rats to stress, the stressor may be perceived as more aversive, but the capacity to adapt to the acoustic stimuli is not impaired. These behavioral effects associated with low serotonin function are likely mediated by specific serotonin receptor subtypes. In **chapter 3** we demonstrated that a low tryptophan diet in combination with a 5-HT_{1A} receptor agonist (flesinoxan) significantly increased the acoustic startle response (ASR), while animals that received the 5-HT_{2C} receptor agonist Ro600175 had significantly lower responses. The latter observation may be of particular clinical interest in the treatment of affective disorders.

In **chapter 4**, we described that central serotonin stores were also markedly depleted during chronic administration of the SSRI, which is not yet restored within 2 days after cessation of the drug. Thereafter**,** we showed that abrupt drug discontinuation also had functional consequences. Cessation of chronically administered citalopram significantly increased the ASR. Interestingly, the behavioral responses on the fifth day of testing correlated positively with increased 5-HT turnover in the amygdala, a limbic area prominently involved in stress physiology and fear. These experiments indicate that shifts in the balance between 5-HT reuptake and synthesis have behavioral consequences, in particular when the buffer capacity is diminished following long term SSRI exposure.

HPA-axis

The increased ASR observed with the low tryptophan condition could be connected with the observed increased levels of glucocorticoids. To investigate a possible causal relation, we have experimentally increased corticosterone levels in rats **(chapter 5)**. Continuous increased corticosterone levels in combination with immobilization did not increase, but rather decrease the initial reactivity and variability in response to acoustic stimuli compared with control animals. Again habituation was not impaired. Abnormalities in the HPA-axis are often seen in patients suffering from psychiatric disorders that are associated with stress and stressful events, including depression (29;372;373). The question remained whether these HPA-axis disturbances impair stress-coping, and thus lead to psychopathology, or that the alterations in HPA-axis activity merely reflect the increased stress levels resulting from the disorder. Our results indicate that a dysfunctional HPA-axis itself does not prevent habituation to stressful stimuli, but it may influence reactivity and thus the impact of and coping style to a stressful event.

Part II: clinical studies

The serotonergic system and psychopathology

In these chapters, the role of serotonin function in sensitivity to stress and psychopathology was studied in somatically ill patients with a chronically reduced cerebral tryptophan availability, and accordingly low serotonin function (49).

Similar to long-term tryptophan depletion in animals, low tryptophan levels in humans were also associated with high corticosteroid levels **(chapter 6)**. The mechanism of this association is unclear. However, because this association was observed in patients suffering from metastatic carcinoid disease, we speculated that having a carcinoid tumor is a stressful event by itself for which subjects with low cerebral availability of tryptophan may be particularly sensitive.

Tryptophan depletion has been associated with various psychiatric symptoms, such as depression, hostility, aggression, and quarrelsomeness (63;115;116;315;374). In **chapter 7** we tried to delineate those symptoms allegedly related to chronically mild diminished tryptophan availability in the central nervous system, as a result of interferon-alpha treatment. Diminished tryptophan availability predicted impairment in social interaction (frequent quarreling, uncontrolled temper and not feeling accepted), but not increased levels of distress (depression / anxiety) nor somatic complaints.

Symptoms associated with low tryptophan availability have also been reported to interact with the 5-HTTLPR polymorphism of the serotonin transporter (62;66;339). In addition, homozygotes for the short alleles of the 5-HTTLPR seemed more sensitive to stress, as witnessed by an increased ASR (375). Accordingly, we investigated the influence of the 5-HTTLPR triallelic polymorphism on psychopathology in patients with long-lasting tryptophan depletion due to a serotonin producing metastatic carcinoid tumor **(chapter 8)**.

Indeed, an interaction of tryptophan levels and 5-HTTLPR on change in severity of psychopathology was observed for total score on the Symptom Checklist-90 (SCL-90) (320) and five of the eight sub-scales, in particular agoraphobia and interpersonal sensitivity-mistrust. This suggests that the s/s homozygotes are more sensitive to plasma tryptophan levels than carriers of the s/l or l/l genotype.

HPA-axis system

In addition, the relation between HPA-axis function and psychopathology was studied. In **chapter 9** we assessed glucocorticoid receptor (GR) function of lymphocytes with dexamethasone ex-vivo and related this to plasma cortisol levels and Hamilton depression scores. The preliminary results indicated that the severity of depressive symptoms correlated with decreased GR function, and that this association was modified, but not entirely determined by the patient's cortisolemic status. These results, combined with the observed behavioral responses of animals with a dysfunctional HPA-axis due to corticosterone pellet administration, suggest that long-term exposure to high levels of corticosteroids may influence the course of the disorders. Our observation that high and persistent corticosterone levels reduced inter-individual variability in rats might be related to the reduced fluctuations in mood as seen in depressed patients. If so, then variations in corticosteroids may point to a more general capacity of an organism to express a variety of affective states. It is tempting to speculate that GR resistance attenuates random fluctuations of mood and, conversely, maximal GR flexibility facilitates recovery.

GENERAL DISCUSSION AND FUTURE PERSPECTIVES

The present studies confirm the hypothesis that sensitivity to stress can be altered by both the serotonergic system and the HPA-axis, and provide a clue for serotonergic determinants of depression or psychopathology in general.

Low cerebral serotoninergic activity increases 1) the physiological response to stress, as witnessed by increased HPA-axis activity, 2) the behavioral response to stress, as witnessed by the increased startle response to acoustic stimuli in rats, and 3) vulnerability to psychopathology in humans. However, an over-active and thus dysfunctional HPA-axis itself seems to reduce the stress response and variation in behavior, suggesting a delicate balance between the two systems.

In humans, most studies regarding the behavioral consequences of chronic low tryptophan levels have focused on depression. Sometimes, a relation with tryptophan was found (140;323;324), but other papers contradict this (317). More recently, also the well-known side effects of treatment with interferon (IFN), being impulsivity and aggression (297;302;326), have been associated with decreased plasma tryptophan levels (115;116;326). Several explanations can be raised for the lack of consistency between serotonin function and psychiatric symptoms.

The first assumption is that serotonin does not lead to specific behavioral problems, but that the role of serotonin is only modulatory. A decreased serotonin function increases the vulnerability to stress, but the impact of serotonergic changes on psychological and social functioning will also be determined by individual characteristics. For instance, not all individuals are susceptible to tryptophan depletion. During experimentally induced acute tryptophan depletion paradigms, mood mainly decreases in subjects with a personal or family history of depressive disorders (e.g. (63;64;325)). In addition, ATD predominantly causes depressive symptoms in women, while the symptoms in men converge towards aggression (66). Genetic variation is also likely to contribute to the individual variability in response to ATD (345). In individuals with a history of major depressive disorder (MDD), the most severe mood deterioration is associated with the 5-HTTLPR l/l genotype. Conversely, in individuals without a history of MDD, the effect of ATD is less severe and here the s/s phenotype is associated with more pronounced changes in anxiety, punishment sensitivity, impulsivity and motivation rather than mood (for review: (345)). These results show that the effect of tryptophan depletion is modulated by genetic make-up, personal history, and possibly also by environmental factors and coping strategies.

A second explanation could be that serotonin is indeed related to specific symptoms, but that those symptoms are not well recognized in conventional diagnoses. Psychiatric disorders are diagnosed according to the consensus criteria stated by *Diagnostic and Statistical Manual of Mental Disorders* Version IV, 4th edition (DSM-IV; American Psychiatric Association, 1994). The diagnosis Major Depressive Disorder for example requires a persistent sad mood or loss of interest interfering with work or family relations for at least two weeks, and at least four additional symptoms such as significant changes in appetite or body weight, difficulty sleeping or oversleeping, agitation or physical slowing, loss of energy, feelings of worthlessness or inappropriate guilt, difficulty thinking or concentrating, disturbances in sexual desire, or recurrent thought of death or suicide. This type of classification of depression has worked very well to standardize the diagnosis of psychiatric disorders by clinicians, but conceals the underlying etiology. Heterogeneity of depressive symptoms and the highly variable individual course of the disorder both suggest that depression is a syndrome consisting of several disorders with a distinct etiology or even pathophysiology (376;377). This classification problem also arises in research. Depression questionnaires contain items regarding different dimensions of MDD. In scientific research, mostly the sum scores of a questionnaire determine whether someone is regarded as having a depressive episode. Accordingly, a patient can be counted as MDD without fulfilling the key symptoms of low mood or anhedonia. To overcome these diagnostic problems, we tried to disentangle those symptoms that are associated with mild and more chronic tryptophan depletion. Historically, tryptophan depletion is associated with low mood. However, decreased mood is mainly found in acute tryptophan depletion studies, where there is a rapid and strong decrease of plasma tryptophan in contrast to our paradigm where there is a slow and relatively mild decrease (378). We found that decreased TRP availability in

relatively mentally healthy patients was associated with bad temper, frequent quarrelling and not feeling accepted (chapter 7). In carcinoid patients with more long-lasting low tryptophan levels, low tryptophan was related to almost every SCL-90 subscale, but most pronounced with agoraphobia and interpersonal sensitivity-mistrust (chapter 8). We have suggested to cover these and some related symptoms together under the umbrella social interaction (chapter 7), which might in some individuals convert to social dysphoria. Such a symptom complex includes uneasiness of a person in social interactions and social cognition (379), resulting in phobic, impulsive and aggressive behavior. The emerging symptom profile and behavior may depend on gender, previous experiences of a subject and perhaps, personality. This idea also fits well in the behavior modulating function of the brain serotonin system (55). It could be that these symptoms are primarily related to serotonin function, but are incorporated in different psychiatric diagnoses and sub-domains of psychopathology questionnaires. We therefore suggest that these symptoms should be regarded as a specific entity.

Because impaired 5-HT function may lead to increased sensitivity to stress and thus facilitates the development of stress-related (psychiatric) disorders, it is of clinical importance to identify the subjects at risk and to intervene on this stress vulnerability. Clearly, subjects at risk could be carriers of the 5-HTTLPR s/s genotype (11;97), or subjects with transient low tryptophan availability (chapter 6), or a combination of both (chapter 9). However, 5-HT function may also be impaired in relation to SSRI treatment. We have shown that acute SSRI discontinuation induces rapid and long-lasting changes in serotonin metabolism and behavior in rats. Because the behavioral symptoms already appear two days after cessation, symptoms associated with SSRI discontinuation may also precipitate during periods of non-compliance in the regular treatment. We feel that too little attention has been directed to the issue of non-compliance. Because discontinuation symptoms bare some similarity with symptoms of depression (111), they may not be recognized as such. This may lead to misdiagnosis and unnecessary switches in treatment. This could even play a role in the incidental, but not unchallenged reports, on suicide and paradoxical effects associated with SSRI treatment (248;380;381).

In vulnerable subgroups, medication might help to ameliorate sensitivity to stress. With low tryptophan availability, therapeutic interventions such as tryptophan enriched diets (315), or medication with drugs to support cerebral 5-HT functioning could be considered (305;327). In the case of non-compliance with SSRIs, preparations with extended drug release may be of benefit. Specific serotonin receptor agonists may also be of therapeutic use. Promising in this respect are selective 5-HT_{2C} receptor agonists. For instance, the selective 5-HT_{2C} receptor agonist Lorcaserin (202) is currently subject of clinical trials for its alleged anti-obesity properties, but it might also ameliorate the psychiatric side-effects associated with some clinical conditions associated with low tryptophan (e.g. carcinoid tumor, inflammation) and medications (e.g. interferon-α with hepatitis C and various cancers) (115;116;382;383). Finally, because SSRIs act as indirect agonists of both 5-HT_{1A} and

5-HT_{2C} receptors, it can be speculated that co-administration of a 5-HT_{1A} receptor antagonist with an SSRI might decrease the irritability and anxiety associated with the early phase of treatment (see also: (203)).

CONCLUSION

The present thesis indicates that changes in serotonergic activity and HPA-axis activity as a consequence of a disease process, treatment condition or other environmental factors in combination with individual genetic make-up may have behavioral consequences, easily to be misdiagnosed by health professionals. This may lead to unnecessary prescription of antidepressants and inadequate treatment in general. We therefore plead for the inclusion of a separate "serotonergic" symptom profile list in all psychiatric questionnaires, including future incarnations of the DSM.