Stress sensitatie

Programme co-ordinator *Prof.dr. J.M. Koolhaas*
Vakgroep Dierfysiologie, Rijksuniversiteit Groningen

Projects:

- *Stress sensitization and CHR signalling*

Project 806.46.061
Projectleader Prof. dr. F.J.H. Tilders (VUA)
PhD-student Dr. A.S.P. Jansen (1999 – 2002)

- *Stress sensitization: who and when?*

Project 806.46.062
Projectleader Prof. dr. J.M. Koolhaas (RUG)
PhD-student Drs. J.G. de Jong (1998 – 2002)

- *Indices of stress sensitization in pigs*

Project 806.46.063
Projectleader Dr. E.M. van der Beek (WUR)
PhD-student Drs. A.G. Karman (1999- 2003)
Techn. assistent A. Van Kesteren-Buiting (2000 – 2002)

Summary
This programme will focus on stress sensitization as one of the fundamental processes determining the adaptive capacity of animals. Environmental conditions and individual factors affecting stress sensitization, as well as some of the neuroendocrine and neurobiological mechanisms will be studied in rats. The resulting neurobiological indices of stress sensitization will be validated in pigs.

Results

project 806.46.061
Chronic stress can cause protracted alterations in behavioral, autonomic and neuroendocrine stress responses. This can result in enhanced responses and reflects a state of stress sensitization and threatens welfare and wellbeing. Recently, it was found that also short exposure to a stressful event (environmental, immune, drug), can lead to long lasting sensitization of stress responses. Such event-induced long lasting stress sensitization is considered to reflect increased stress vulnerability and plays a role in the development of stress pathology. We hypothesized that neuroplasticity of stress responsive systems in the brain underlies acquired stress vulnerability. The goal of this project was to describe these adaptive alterations in the brain.

Because corticotropin releasing hormone (CRH) signaling in the brain, is known to facilitate behavioral, neuroendocrine and autonomic stress responses, we tested the hypothesis that altered CRH signaling plays a role in stress sensitization. Using a short “immune stress” in adult rats as a model, we found that CRH gene expression and peptide storage is enhanced in hypothalamic CRH neurons 3 weeks later i.e. during the peak of the neuroendocrine stress sensitization. In addition, we found that hypothalamic CRH neurons up-regulate the expression levels of the CRH type 1 receptor (CRHr1), that play a role in auto-excitation of these neurons.

Taken together these data suggest that indeed increased hypothalamic CRH signaling contributes to stress sensitization. In order to find out whether changes in CRH receptors occur throughout the brain, we used CRH receptor autoradiography of brain slices, involving 125I-sauvagine as a ligand and the CRHr1 antagonist R121919, to collect specific information on type 1 and 2 CRH receptors. By using an image analysis system to measure CRH receptor binding in defined brain structures, we found heterogeneous distribution of CRHr1 and CRHr2 in the brain but no stress-sensitization associated

alterations in CRHr1 and CRHr2 binding in any of the brain areas studied. We conclude that altered CRH signaling is either restricted to the hypothalamus or not associated with major alterations in CRH receptors.

Therefore we shifted attention to another stress system on which we had a promising lead. Because amphetamine-induced stress sensitization was not associated with neuroplastic changes in hypothalamic CRH neurons as seen during immune-induced sensitization, we hypothesized that functional alterations in neurons that drive the hypothalamic CRH system during stress may play a role in stress sensitization. Indeed, our data reveal that both drug- (amphetamine) and immune-(interleukin-1) induced sensitization is associated with enhanced electrically evoked release of noradrenaline in the paraventricular nucleus of the hypothalamus (PVN), where CRH neurons are localized. To study whether these functional alterations are associated with restructuring of the innervation density or pattern, we used an image analysis approach of brain sections stained for dopamine-beta-hydroxylase as a label for noradrenergic nerve fibers. Our findings show that both interleukin-1 and amphetamine induce plastic changes in the noradrenergic innervation density in the PVN, in particularly in those parts of this hypothalamic nucleus that are rich in CRH neurons.

Novel scientific questions

Questions with respect to pathological alterations in CRH signaling associated with stress-vulnerability and associated pathology are presently tested by various research groups and in patient populations using CRH receptor antagonists.

The decline in the innervation density by specific populations of noradrenergic neurons that project to the PVN presents an apparent paradox with the increased noradrenaline release in this nucleus, that needs to be elucidated. It may relate to rearrangement of excitatory and inhibitory noradrenergic input to this nucleus, to the advantage of excitatory input.

Whether alterations in connectivity between noradrenergic neurons and CRH neurons play an instrumental role acquired vulnerability to stress needs to be studied in other models and pathologies. The studies will be continued with support from other sources.

Publications

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- Schmidt, ED, Aguilera, G, Binnekade, R, Tilders, FJH . Single administration of interleukin-1 increased corticotropin releasing hormone(CRH) and CRH-receptor mRNA in the hypothalamic paraventricular nucleus which paralleled long lasting sensitization to emotional stressors (submitted)

Project 806.46.062

Differential vulnerability of proactive and reactive coping male rats to the long-term effects of social defeat

In this project the influence of individual differences in coping strategies on the long-term effects of a single social defeat was investigated. Animals may adopt either a proactive or reactive coping strategy. These two behavioral styles differ not only in their behavioral and neuroendocrine response to stressors, but show also differences in several neurotransmitter systems.

From an randomly bred strain of wild-type rats, animals were selected with either a high or low aggression level. The tendency to initiate aggressive behavior has been shown to be predictive for the individual reaction to other environmental challenges. Animals were then subjected to a social defeat by a dominant rat and changes in behavior and physiology were studied for a period of four weeks. Using a radio-telemetry system, heart rate, temperature, and activity were recorded in the home cage and reactivity to mild stressors was determined. Preliminary results point to an important role of individual variation in coping strategies in determining the long-term consequences of social defeat. Both high and low aggressive animals show long-term changes, but these are expressed in different parameters.

Stress sensitization and serotonergic transmission

Previous research has shown that in Wistar rats the adrenocortical response to a challenge with 8-OH-DPAT, a 5-HT1A receptor agonist, is reduced shortly after social defeat, indicative of a lower sensitivity of the postsynaptic 5-HT1A receptor. When the long-term effects of defeat on the serotonergic regulation of the HPA-axis were studied in wild-type rats, desensitization of the adrenocortical response proved difficult to measure due to the high variation in baseline adrenocortical activity.

In a new experiment, again using Wistar rats, short- and long-term effects of social defeat on the efficacy of the postsynaptic 5-HT1A receptor were determined using the 8-OH-DPAT induced hypothermic response. This response was determined via a radio-telemetry system 7 days before and either 1 or 21 days after social defeat. Results show a clear desensitization of the postsynaptic 5-HT1A receptor on day 1 after defeat, but not on day 21.

Stress sensitization and dopaminergic neurotransmission

Chronic stress can cause cross-sensitization for amphetamine. Previous experiment showed that also a single social defeat can produce an enhanced locomotor response to amphetamine in an open field. This effect occurred only on day three after defeat, but not on days 14 and 21.

In a subsequent experiment cross-sensitization was measured in the home-cage to exclude possible effects of novelty. Animals were equipped with transmitters to measure heart-rate, temperature and activity and then exposed to either social defeat, a high dose of amphetamine or a control treatment. Injection with a challenge dose of amphetamine was performed 3, 7 and 21 days after treatment. Results show a clear sensitization in the amphetamine treated group, which was relatively long lasting. Sensitization after social defeat was observed 3 days after defeat, and again appears to be of short duration.

Publications

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- De Jong, Meijering & Koolhaas: Single social defeat causes long-term desensitization to the hypothermic effects of amphetamine (in prep.).

- De Jong & Koolhaas: Individual variation in behavior determines the long-term effects of social defeat (in prep.).
- De Jong & Koolhaas: Housing conditions determine the long-term effects of social defeat (in prep.).

Project 806.46.063

Young male and female pigs were group housed under standard impoverished (control: concrete) or slightly enriched (rich) concrete + straw bedding) conditions from weaning until 26 weeks of age. All animals were characterised on days 3 and 10 after birth by means of the back test as high- or low-resistant. We found no significant differences in the expression of vasopressin (VP) as determined by immunocytochemistry in the paraventricular nucleus of these animals between type or housing conditions. Also, no effect of type or housing condition was found on the amount of immunoreactive corticotropin releasing hormone (CRH). Yet, control housed animals showed a significant correlation between the amount of VP expression and CRH expression in the hypothalamus. This positive correlation may indicate an adaptation of the reactivity of the HPA-axis under poor housing conditions. Furthermore, VP expression differed significantly between sexes, i.e. VP content of the PVN was significantly lower in females compared to males, irrespective of housing condition.

In a first study in group and individual -housed 1 year old female virgin pigs (gilts), we found a significant effect of individual housing on the VP expression in high resistant (HR), but not in low resistant (LR) classified animals. Further research will focus on the expression of CRH in these animals.
To study mRNA expression of CRH in the hypothalamus of these pigs, a non-radioactive ISH technique was set up. This technique will be used to study possible effects of housing conditions such as group and individual housing on CRH and VP expression in the hypothalamus. This dual ISH-immunocytochemical approach could give us insight in the relation between gene expression, peptide content and behavioural characteristics that are influenced by the hypothalamus-pituitary-adrenal axis.

Publications

Abstracts

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Papers

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- artikel resultaten in VP en CRH in group housed animals, effect van verrijking
- artikel resultaten VP in gelten, effect van huisvesting (groep versus individueel)
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