

Corticotropin-releasing hormone (CRH) plays a pivotal role in the response of an organism to various stressors. Furthermore, chronic elevated levels of CRH are implicated in human stress-related and affective disorders, including major depression. To gain more insight into the relationship between hyperactivity of the CRH system and associated neuroendocrine, autonomic, physiological and behavioral changes, we have developed a transgenic mouse model of lifelong CRH overproduction (CRH-OE). In this study, we explored the behavioral consequences of chronic CRH overproduction in mice of two established transgenic lines (CRH-OE2122 and CRH-OE2123) in behavioral paradigms reflecting different aspects of stress, anxiety and depression. These paradigms include the open field test, which is based on free exploration of an unfamiliar environment, the elevated plus maze, which is associated with anxiety-like responses, and the forced swim test, which can detect (anti-)depressant-like behaviors.

MATERIAL & METHODS

The generation of the transgenic CRH overexpressing mice is described extensively elsewhere [1]. Male transgenic mice were used in these experiments. Littermate wildtype (WT) mice served as controls. The mice were 11-16 weeks old at the time of testing.

Spontaneous locomotor behavior was quantified in an open field. The open field was a round, opaque plastic box (diameter 45 cm, height 29.5 cm), divided into a center, middle and outer ring, placed in a sound-attenuated box to allow recording of undisturbed behavior (illumination approx. 2400 lux). The behavior of the mouse was recorded over a 10-min period using EthoVision (detection method: subtraction absolute, sample rate 5 samples/s, noise threshold 30, image filter erosion/dilation 1 pixel, contrast 48).

The elevated plus maze was made of black Plexiglas with arms 29 cm long and 5 cm wide, extending from a central platform (5 cm square). Two opposite arms were enclosed by black Plexiglas walls (15 cm high; illumination level 30 lux) and two arms were open (illumination level 60 lux) with a small ledge (0.25 cm high). The mouse was placed on the central platform facing an enclosed arm and its behavior was recorded over a 5-min period using EthoVision (detection method: subtraction absolute, sample rate 3 samples/s, image filter erosion/dilation 1 pixel, contrast 45).

For the forced swim test, clear plastic cylinders (diameter 12 cm, height 25 cm) were filled to a depth of 10 cm with water (25 °C). Mice were placed in the water for 6 min and their behavior was recorded on video. The same procedure was repeated the following day. Videotapes were analyzed using The Observer, and the durations of immobility, swimming and climbing were scored only during the last 3 min.

RESULTS

Locomotion in a novel open field, more specifically in the outer perimeter of the open field, was significantly reduced in CRH-OE2122 mice, but there was no difference between CRH-OE2123 and WT mice. In the elevated plus maze, overexpression of CRH was not associated with changes in behavior indicative of altered anxiety levels, as evidenced by the similar percentages of time spent on the open arms and the similar percentages of open arm entries across genotypes. In the forced swim test, all genotypes exhibited similar amounts of immobility. Furthermore, there were no genotype differences in the duration of climbing behavior, though differences in swimming time nearly reached significance.

CONCLUSIONS

The Observer and EthoVision proved to be very useful tools in this study, in that they allowed easy recording and analysis of specific behaviors, resulting in an extensive description of the behavioral profile of the mutant mice overexpressing CRH. In addition, both programs yielded reliable data for statistical analysis. The only relatively consistent finding, although not always significant, was reduced locomotor activity in CRH-OE2122 mice, corresponding well with the known effects of CRH on locomotion. Contrary to our predictions, the results of the present experiments with CRH-OE2122 mice indicate that lifelong CRH overproduction is not associated with an apparent behavioral phenotype indicative of enhanced stress, anxiety and/or depression.

REFERENCES

1. Dirks, A.; Groenink, L.; Lutje Schipholt, M.; van der Gugten, J.; Hijzen, T.H.; Geyer, M.A.; Olivier, B. (2002). Reduced startle reactivity and plasticity in transgenic mice overexpressing corticotropin-releasing hormone. *Biological Psychiatry*, **51**, 583-590.

CONTACT INFORMATION

Anneloes Dirks. Department of Psychopharmacology, Utrecht University, Utrecht, the Netherlands.

E-mail: a.dirks@pharm.uu.nl