Prenatal stress produces anxiety- and depression-related behavior particularly in male Sprague-Dawley rats

D.L.A. Van den Hove[1*], G. Kenis[1*], M. Bruschettini[1], C.E. Blanco[2], H.W.M. Steinbusch[1], J. Prickaerts[1]

[1]*Department of Psychiatry and Neuropsychology, Research Institute Brain and Behavior, Division Cellular Neuroscience, Faculty of Medicine, European Graduate School of Neuroscience (EURON), Maastricht, The Netherlands {d.vandenhove@np.unimaas.nl}*

[2]*Department of Pediatrics, Research Institute Growth and Development, Faculty of Medicine, Maastricht University, Maastricht, The Netherlands*

**These authors contributed equally to this work*

Background

Nowadays, it has become increasingly clear that adverse events experienced by a pregnant woman, be they physical or emotional, may be reflected upon the developing fetus and adversely affect its physical and mental wellbeing in later life. In the present study we examined the effects of prenatal restraint stress on anxiety- and depression-related behavior in both male and female Sprague-Dawley rats at 4-5 months of age.

Behavioral assessment

Anxiety-related behavior was evaluated in the elevated zero maze and the home cage emergence test. Depression-related behavior was examined using the forced swim test

The elevated zero maze consisted of a circular alley (diameter of 100 cm; path width 10 cm) made from black plastic material that was transparent for infrared light and elevated 20 cm above the floor. The maze was divided in four parts, i.e., two opposite open parts and two opposite closed parts with sidewalls 30 cm height. The open parts had borders with a height of 5 mm to prevent the rat from stepping down from the apparatus. For the test, the rat was placed into one of the open parts facing a closed part of the apparatus. After five minutes the rat was removed from the apparatus and the maze was cleaned with ethanol and water and dried thoroughly. The movements of the rat were scored automatically under dark conditions with a computerized system using an infrared video camera (EthoVision Pro, Noldus, The Netherlands). Percentage of time spent in the open part of the maze and total distance traveled were determined.

For the home cage emergence test the rat's home cage (opened) was placed in the center of an open field ($1m^2$) and the rat was allowed to leave its cage via a grid walkway. The latency to emerge from the home cage (i.e., four paws on the grid) was scored. If the rat did not emerge from its home cage within 300 sec, the session was ended, the home cage was closed again and the rat was given a score of 300 sec. This test was carried out on 3 consecutive days under low light conditions.

In the forced swimming test four cylindrical glass tanks (50 cm tall, 20 cm in diameter) were filled to a height of 30 cm with 25°C water. The movements of the rat were scored automatically with a computerized system (EthoVision Pro, Noldus, The Netherlands) during a 5 min session under low light conditions. Scored were 'immobility' which reflects no movement at all and/or minor movements necessary to keep the nose above the water) and 'strong mobility' reflecting 'escape behavior' (e.g. climbing against the walls and diving). Settings within EthoVision were adjusted based on manually recorded sessions and were attuned for each gender separately (immobility/mobility threshold: 12 and 20; mobility/strong mobility threshold: 16.5 and 23.9 for males and females, respectively).

Results

Prenatal stress was associated with a clear increase in anxiety-related behavior in male, but not female offspring, as evaluated in the elevated zero maze and the home cage emergence test. Likewise, depression-related behavior in the forced swim test was increased in prenatally stressed male rats only. Prenatally stressed male offspring further showed increased basal plasma corticosterone levels, whereas both prenatally stressed males and females failed to show an adequate response to stress with lower stress-induced corticosterone levels as compared to controls. Female hippocampal weight was relatively higher after prenatal stress, which may explain the absence of clear behavioral effects of prenatal stress in this gender. In addition, male birth weight was a predictive marker both for performance in the forced swim test, as well as for plasma corticosterone levels in adulthood.

Discussion

In conclusion, prenatal stress resulted in increased anxiety- and depression-related behavior particularly in male Sprague-Dawley rats. Females seemed to be relatively protected towards the effects of prenatal stress.