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Influence of sexual experience on olfactory sensory activity in mice.

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Abstract

Male mice undergo a striking behavioral switch after sexual experience: virgin males are infanticidal, but after sexual contact with a female they change their behavior towards the young, becoming non-aggressive, parenting individuals. In this project, our goal was to investigate the changes in olfactory sensory activity that accompany these behavioral changes. We used immunostaining to measure the activity levels in sensory neurons of one of the olfactory organs in the nasal cavity of mice, known as the vomeronasal organ. This approach was performed in virgin male mice and in males after varying periods of sexual contact and cohabitation with a female. As a result, we were able to correlate the olfactory sensory activity with the behavior in the same animal groups, and it became clear that, as mice become progressively less aggressive during the switch, the sensory activity decreases.

Key words:

Neurobiology, olfactory organ, infanticidal behavior.

Introduction

In mammals, important sensory information is detected by the olfactory sensory system, which includes the vomeronasal organ (VNO). The VNO mediates a range of instinctive behavioral responses, like male-male aggression and parental care (Dulac & Torello, 2003)

Aggressive behavior towards newborns is exhibited by virgin males, but is not present in sexually experienced adults (vom Saal & Howard, 1982). Previous data show that, after mating and cohabitating with a female, the activity of the vomeronasal system decreases in male mice (Tachikawa et al, 2013). These data suggest that sexual experience modulates the behavior towards pups through changes in olfactory activity.

To investigate the role of sexual experience in modulating the neuronal VNO activity, we used immunostaining to assay the expression of an indirect marker of neuronal activity, pS6, on VNO histological sections from males in experimental groups defined by the presence and duration of sexual contact with females.

Results and Discussion

We analyzed male mice in 5 groups: virgin males, dpc2 (2 days of sexual contact and cohabitation with a female), dpc7, dpc15 and dpc22. The vomeronasal organs were collected from these animals (n=3 individuals per group), fixed in 4% paraformaldehyde and cryoprotected prior to sectioning on a cryostat to produce 16µm histological sections. Sections on glass slides were subjected to immunostaining with an antibody against pS6, the phosphorylated version of ribosomal protein S6, an indirect marker of neuronal activation in olfactory organs.

This approach allowed us to investigate how VNO activity changes in male mice, influenced by the sexual experience and cohabitation with a female. We also performed behavioral assays on animals in the same groups, in order to determine the correlation between olfactory sensory activity and the exhibited behavior. Importantly, we analyzed infanticide behavior, defined as the percentage of adult males in each group that exhibit aggressive behavior towards a 3-days old pup in a 1 hour assay. It should be noted that during the assay, the pups

were only fully exposed for 3 minutes, and then protected in a special protection cage.

The results obtained from this experiments show that a major decrease in both vomeronasal neuronal activity and behavior, happens after 7 days of sexual contact and cohabitation with a female and is sustained afterwards. Although the decrease in VNO activation of parental mice has been previously described, our data show a temporal series of such decreasing and its correlation with the infanticidal behavior, that hasn't been previously studied.

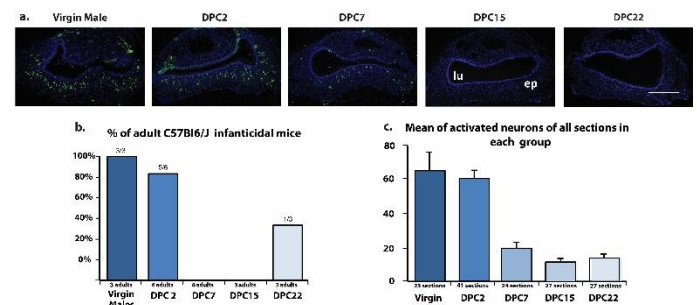


Figure 2: Representation of the obtained results. (a.) fluorescence microscopy images of VNO tissues after immunostaining experiment. Every green dot is an activated neuron. Lumen (lu) and sensory epithelium (ep) are represented and the white bar equals 100µm. **(b.)** Percentage of infanticidal adult mice in each group. Above the bars, the number of infanticidal male mice adults by the totality of exposed adults in each group is shown. **(c.)** Representation of the means of activated neurons counted in all the sections obtained for each group.

Conclusions

Our data show that the behavioral switch in male mice after sexual experience with females is accompanied by a marked decrease in activity in the olfactory vomeronasal organ. In the future, we intend to investigate the causal relationship between the behavior and sensory activity and the molecular mechanisms underlying such phenomenon.

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