Medial Amygdala Lesions in Male Rats Reduce Aggressive Behavior: Interference With Experience

J D VOCHTELOO AND J M KOOLHAAS

Department of Animal Physiology, State University Groningen
P O Box 14, 9750 AA Haren, The Netherlands

Received 13 April 1987

VOCHTELOO, J D AND J M KOOLHAAS Medial amygdala lesions in male rats reduce aggressive behavior. Interference with experience. PHYSIOL BEHAV 41(2) 99-102, 1987 — The medial nucleus of the amygdala (am) has been implicated in a variety of social behaviors. The present experiment will test the hypothesis that the effect of am lesions on intermale aggressive behavior is due to interference with social learning processes. Small electrolytic lesions of the am had no significant effect in socially naive male rats. A marked reduction of aggressive behavior was observed in animals that were lesioned after experience with four aggressive interactions. Repeated testing suggests that these lesioned animals are unable to adapt their aggressive behavior on the basis of previous victory experience. The results are discussed in terms of social learning and memory processes in relation to the afferent and efferent connections of the am.

Medial amygdalasqlater aggressive behavior Learning processes

The present paper will consider the possibility that the changes in aggressive behavior observed by some authors after amygdala lesions may be due to interference with social learning processes. This suggestion was also made by Bolhuis et al [1] on the basis of deficits after cortico-medial amygdala lesions in the conditioned avoidance of a dominant observed in experimental animals that have been defeated by this dominant male. Considering the afferent and efferent connections of the medial amygdala, Luiten et al [15] argued that this part of the amygdaloid complex might be quite specifically involved in social behavior. By testing the effects of medial amygdala lesions in intermale territorial aggressive behavior, the present experiment will test the possibility that the medial amygdala is involved in this type of social behavior. Moreover, by using groups of experimental animals that differ in the degree of experience with intermale aggression we can test the hypothesis that the effects on this type of behavior are due to interference with social learning processes.

METHOD

Experimental Animals

Fifty-one male rats (Tryon Maze Dull S3) of 6 months of age were used. They were housed individually in wooden observation cages (80×55×50 cm) together with a female that was sterilized by ligation of the oviducts. Food and water were available ad lib. The cages were placed in a temperature controlled room (20°C) with a light-dark cycle of 12.
Fig. 1. Coronal sections of the rat brain showing the total area covered by the lesion (N=21).

hours light and 12 hours dark. All experiments were performed during the first half of the dark period.

Stimulus Animals

To elicit territorial aggressive behavior by the experimental animals, male rats (Tryon Maze Dull SR) were used as intruders into the home cage of the experimental males. These animals were housed in groups of 4 animals in cages (55×30×15 cm) with ad lib food and water. All intruder rats were about 50 grams less in body weight than the experimental animals. In order to obtain a standard stimulus animal, all intruders were three times confronted and defeated by a trained fighter rat before they were used in the experimental procedure. Each stimulus rat was confronted with the same experimental rat only once.

Surgery

At the time of surgery the animals were anesthetized with ether and placed into a stereotaxic apparatus (David-Kopf). Bilateral lesions were made with a monopolar stainless steel electrode with a diameter of 0.2 mm and a uninsulated tip of 0.1 mm. The electrode was aimed at the medial amygdala (coordinates anterior 5.3, lateral 3.5 and ventral 8.6 mm below dura) [18]. Lesions were made with an anodal current of 1.2 mA during 5 seconds. In the sham operated controls the electrode was lowered 7.6 mm below dura and no current was used.

Experimental Procedure

The group of experimental animals was split into two subgroups. One group of 32 animals consisted of 9 sham lesioned controls and 23 animals that were lesioned before any behavioral testing took place (inexperienced). The other group was either lesioned (10 animals) or sham lesioned (9 animals) after experience with 4 tests for aggressive behavior (experienced). To test the effect of the medial amygdala lesion, each group was tested during three consecutive days, starting 8 days after surgery.

Behavioral Tests

The experimental animals were tested for intermale, territorial aggressive behavior. The behavioral test started at the moment a stimulus rat was introduced into the home cage of the experimental animal. Each test lasted 10 minutes. The duration of the following behavioral elements was recorded by means of a keyboard operated microprocessor: investigate genital sniff, fixate, lateral threat, keep down, offensive upright, clinch, chase (see also [14]). Fixate is characterized by piloerection, both eyes are half closed and the head of the animal is close to the substrate and oriented towards the intruder. Clinch is the fight itself during which the two animals rapidly roll and jump in close contact, biting may occur. The sum of clinch, chase, keep down, offensive upright and lateral threat was used as a composite score for offensive behavior. Attack latency is defined as the time between start of the test and the first clinch. All behavioral measures are expressed as percentage of the total test duration.

Histology

At the completion of behavioral testing, the lesioned animals were deeply anesthetized with sodium pentobarbital and perfused with saline followed by a 4% formaldehyde solution. Frozen sections of 40 μm were cut and stained with cresyl violet for histological examination. The degree of medial amygdaloid damage was determined by measuring the surface in square millimeters of the bilateral medial amygdala lesion per median section of the stereotaxic atlas of Pellegrino and Cushman [18].

Statistics

The Wilcoxon-matched pairs signed rank test was used for comparison within one group of experimental animals. For between group comparisons the Mann-Whitney U test was used [21]. To determine possible effects of the sham lesion on repeated testing, a 2 way ANOVA was used [23].

RESULTS

Twelve lesioned animals were excluded from further analyses because of a unilateral medial amygdala lesion. Twelve of the 21 successfully lesioned animals belonged to the experienced group (mean medial amygdala damage 34.9% SD±15.6) and nine to the inexperienced one (mean medial amygdala damage 27.7% SD±6.2). Since there is no statistical difference in lesion size between the two groups of
experimental animals ($p>0.10$), the total area covered by the lesion in all 21 experimental animals is given in Fig. 1. As can be seen, the lesion caused little or no damage to other amygdaloid structures.

The effects of the lesion on total offensive behavior is shown in Fig. 2. In the inexperienced group, there is a small, non-significant reduction in the relative time spent on offensive behaviors. In the experienced group, however, the lesion caused a significant ($p<0.01$) decrease in offensive behavior, both in comparison to the prelesion level and the sham operated controls. This reduction in offensive behavior in the experienced group appeared to be significantly correlated (Spearman $r=0.66$, $0.01<p<0.05$) with the size of the medial amygdala lesion.

In order to appreciate in more detail the possible interference of the lesion with experiential factors, the effect of the lesion is analyzed on various components of offensive behavior in the course of the test days. Figure 3 shows the relative time spent on clinch and chase, threatening and the attack latency time per test in the two groups of experimental animals. A two-way analysis of variance reveals a significant lesion effect in the experienced group in clinch and chase ($p<0.01$) and attack latency ($p<0.05$) and a weakly significant lesion effect in threatening ($p<0.10$). No significant effects were found on the test day variable. Thus, in contrast to the results in the inexperienced group of experimental animals, there is a significant test day effect both for clinch and chase ($p<0.01$) and the attack latency ($p<0.01$), whereas no significant results were found on the treatment variable ($p>0.10$).

**DISCUSSION**

The results show that small bilateral electrolytic lesions in the medial nucleus of the amygdala reduces male offensive behavior. This reduction is most prominent, and reaches significance only in the experienced group of experimental animals. It is important to notice that the offensive behavior is not totally abolished by the lesion. The lesioned animals are still offensive and do win fights, but at a much lower frequency than their sham operated, experienced controls. This suggests that only a modulating component of the behavior is affected by the lesion. The very marked reduction in offensive behavior in the experienced group suggests that this might be due to a learning and memory component. Since very little is known about the role of learning processes in offensive behavior, it is hard to be more explicit about such a conclusion. Analysis of the sham operated controls reveals that the strongest changes in offensive behavior can be observed during the first three confrontations of inexperienced animals with an unfamiliar opponent. After this initial change, the level of offensive behavior seems to stabilize. The most significant alterations due to the lesion were found when the lesion was placed after this initial period of changes, i.e., in the experienced group. Moreover, offensive behavior or its components remained at a stable low level despite repeated testing. This is in contrast to the effects of a medial amygdala lesion in inexperienced animals. None of the offensive behavioral elements are significantly affected by the lesion, not even after repeated testing, although there may be a small effect in clinch and chase.

The strong and consistent reduction of offensive behavior in the group of animals that were lesioned after four experiences with an intruder into their home cage may be interpreted in terms of learning processes or more specifically retrieval processes. This interpretation is in agreement with the conclusion based upon the avoidance of a dominant [1]. Here too, a medial amygdala lesion results in a behavioral (avoidance) deficit, but only when the lesion is placed after the acquisition of a defeat [1]. It is not easy to explain why there are still no differences between lesioned animals and controls after repeated testing in the inexperienced group. Interestingly, Ekelander et al. [6] found deficits in shock avoidance learning after amygdala lesions in adult animals and no deficits when the adult experimental animals were amygdala lesioned as a juvenile. Hence, it seems as if in the naïve animal several alternative learning mechanisms are still open, that might disappear after repeated experience.

Whatever the exact role of the medial amygdala may be, the results show that this part of the amygdaloid complex is involved in intermale aggressive behavior. The positive correlation between size of the medial amygdaloid damage and the reduction in aggression indicates that the effect is specifically due to the medial amygdala. Several authors emphasized the role of the medial nucleus in social behaviors including male sexual behavior [8,9] and social avoidance behavior [1]. Indeed, the medial amygdala seems to be part of a neural circuitry involved in social behavior in general [15]. Moreover, it is the only amygdaloid nucleus that contains testosterone sensitive neurons [24]. In addition, androgen dependent vasopressinergic cell bodies and terminals related to sexual differentiation [4] have also been described. Hence, the reason for the specific involvement of the medial
nucleus in social behavior seems to be its specific sensory input of both internal (androgens) and external (vomeronasal) origin, and its efferent connections to neural structures such as the ventromedial hypothalamus, the central gray and the preoptic area, reported to be involved in social behavior as well.

As to the function of the medial amygdala, the present results indicate that this nucleus may be involved in the modulation of social behavior on the basis of past social experience whether it is victory or defeat.

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