Title page

Title: Neural mechanisms of predatory aggression in rats - implications for abnormal intraspecific aggression

Running title: Neural background of predatory aggression

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Abstract

Our recent studies showed that brain areas that are activated in a model of escalated aggression overlap with those that promote predatory aggression in cats. This finding raised the interesting possibility that the brain mechanisms that control certain types of abnormal aggression include those involved in predation. However, the mechanisms of predatory aggression are poorly known in rats, a species that is in many respects different from cats. To get more insights into such mechanisms, here we studied the brain activation patterns associated with spontaneous muricide in rats. Subjects not exposed to mice, and those which did not show muricide were used as controls. We found that muricide increased the activation of the central and basolateral amygdala, and lateral hypothalamus as compared to both controls; in addition, a ventral shift in periaqueductal gray activation was observed. Interestingly, these are the brain regions from where predatory aggression can be elicited, or enhanced by electrical stimulation in cats. The analysis of more than 10 other brain regions showed that brain areas that inhibited (or were neutral to) cat predatory aggression were not affected by muricide. Brain activation patterns partly overlapped with those seen earlier in the cockroach hunting model of rat predatory aggression, and were highly similar with those observed in the glucocorticoid dysfunction model of escalated aggression. These findings show that the brain mechanisms underlying predation are evolutionarily conservative, and indirectly support our earlier assumption regarding the involvement of predation-related brain mechanisms in certain forms of escalated social aggression in rats.

Keywords: muricide; aggression; c-Fos immunohistochemistry; hypothalamus; amygdala; periaqueductal grey

1. Introduction

The general functions of aggression fall into two main categories, particularly social competition and predation [1-3]. The first entails conspecifics, which fight for access to resources in a broad meaning (e.g. food, territory, social rank, etc.). This form of aggression is associated with high physiological arousal, and covers social communication. E.g. threat signals convey information on 'attack intentions'; moreover, threats may replace actual fights by the process of ritualization. In contrast, predation aims at killing an individual that belongs to a different species. This behavior is associated with minimal arousal, and does not involve social communication. These two forms of aggression are controlled by distinct neural circuits as shown by feline stimulation studies [4-6]. Based on phenomenological and physiological similarities, these types of aggressive behavior were proposed to be analogous with particular forms of psychopathological human aggression [7-10]. E.g. exacerbated affective aggression is seen in intermittent explosive disorder, which is a violent response to a perceived threat. Other forms of pathological aggression, e.g. those seen in antisocial personality disorder, have different characteristics: such aggression is often gain-oriented, and is associated with limited emotional arousal and low intention signaling [7-9]. The phrase 'predatory aggression' is frequently used to emphasize these characteristics [11-13].

In recent years, the differentiation of the two types of aggression and the idea that they are governed by distinct neural mechanisms gained attention in both human and animal research. In humans, a psychiatric inventory was developed to differentiate reactive and proactive aggression [14], and current theories deal with their neural underpinnings differentially [15, 16]. We recently developed two laboratory models that mimic important characteristics of affective/reactive and instrumental/proactive forms of aggression, and proposed behavioral methods to differentiate species-typical and abnormal forms of aggression [17-20]. Importantly for the present study, we found that rats submitted to one of

these models –the glucocorticoid dysfunction model of abnormal aggression– deliver bites to vulnerable body parts of conspecifics (head, throat and belly), which is accompanied by low intention signaling by threats, disturbed social behavior, and reduced autonomic arousal, features that are in many respects to similar to the symptoms of aggressive antisocial personality disordered subjects [17-20]. We found that in this model, aggressive encounters increase the activation of the lateral hypothalamus, central amygdala and ventral periaqueductal grey (PAG) above the levels seen in controls submitted to fights (i.e. these regions were overactivated) [4, 18, 19, 21, 22]. Moreover, the activation of the central amygdala and lateral hypothalamus correlated significantly with the share of abnormal, predatory-like attacks in this model [22]. As the very same brain regions were shown to control predatory attacks in cats [1, 4-6], we proposed that antisocial-like aggressiveness in rats has a 'predatory dimension' as it regards both behavior and brain function. Unfortunately, however, the brain mechanisms of predatory aggression are less well known in rats than in cats. Early electrophysiological studies identified the hypothalamic locus of control of frog and mouse killing in rats but in contrast to cats, such studies provided limited information on other modulatory brain regions [23-26]. More recently, neural mechanisms were evaluated in rats by using c-Fos immunohistochemistry to investigate brain activation patterns of cockroach hunting as a model of predatory aggression [27, 28]. In these studies, food intake inherently associated with cockroach hunting was carefully controlled. While the activated brain areas overlapped in many respects with the circuitry that controls predation in cats, important differences were also observed. E.g. the lateral hypothalamus showed similar levels of activation in insect hunting rats and their feeding controls, despite the fact that this brain area is considered the most important control region of rat killing in cats and frog and mouse killing in rats [1, 4, 29]. According to our own observation, muricide and insect hunting are behaviorally different, which may explain these discrepancies. As such, studies using

muricide as a model seem necessary to fully understand the brain mechanisms of predatory aggression in rats, but such studies are unavailable at present.

Here we investigated c-Fos activation in 15 aggression-related brain regions in adult male Wistar rats that spontaneously killed a mouse in their home-cage. Rats which did not attack the mice and rats without mouse exposure were used as controls. This study was motivated by multiple goals. Firstly, we aimed at describing brain activation patterns associated with muricide, a work that has not been performed so far. Secondly, we aimed at comparing these findings with those obtained earlier in cat stimulation studies to establish the cross-species stability of predation-related brain mechanisms. We also aimed at comparing findings with those obtained in the cockroach-hunting model, to investigate the impact of the pray on brain mechanisms. Finally, we aimed at providing a more proper comparison for the recently described "predatory-like aggression network" activated in the aforementioned model of violent social aggression [22].

2. Material and methods

2.1. Animals

Subjects were adult male Wistar rats raised in the breeding facility of our Institute. Parents were obtained from Charles River Laboratories (Germany). Rats were housed in macrolon cages in groups of 4-6. Food and water were available *ad libitum* throughout, temperature and relative humidity was kept at 22±2 ºC and 60±10 %, respectively. Rats were maintained in a light cycle of 12:12 hours with lights off at 1000h. The weight of subjects was 350-450 g when behaviorally tested. Behavioral tests were conducted in the early phase of the dark period, under dim red illumination. 50-70 days-old male CD1 mice from the same source were used as stimulus animals. Mice were housed in a different room, but otherwise were maintained under similar conditions. The experiments were carried out in accordance

with the European Communities Council Directive of 24 November 1986 (86/609/EEC) and were reviewed and approved by the Animal Welfare Committee of the Institute of Experimental Medicine.

2.2. Experimental procedure

Subjects were housed individually for one week before behavioral testing but otherwise were maintained under the same conditions as earlier. The experiment was started by placing a mouse in the home-cage of the rat. Subjects have never encountered a mouse before. If the rat killed the mouse, the latency to kill was recorded, the killed mouse was removed immediately, and at the same time, another, uninjured mouse was removed from the home cage of a randomly chosen rat to assure that the time of stimulation/interaction was equal between groups ("muricide" vs. "no muricide" control). The cut-off time for mouse-killing was 20 min. On each experimental day, control rats unexposed to mice („no mouse" control) were also randomly chosen from rats that were not exposed to mice to assess baseline c-Fos activity. The experiment was continued until sample sizes reached 7 per group.

2.3. Brain processing and immunohistochemistry

Rats were left undisturbed for 120 minutes after the encounters to allow c-Fos signal to develop. Afterwards, they were deeply anesthetized by an i.p. injection of a mixture of ketamine, xylazine and pipolphen (5, 10 and 5 mg/kg, respectively) and perfused through the ascending aorta with 100 ml ice-cold 0.1M phosphate-buffered saline followed by approximately 200 ml 4% paraformaldehyde dissolved in 0.1M phosphate-buffered saline. The brains were removed, post-fixed in the same solution for 3h and cryoprotected overnight by 20% sucrose in phosphate-buffered saline at 4 ºC. 30μm frozen sections were cut in the frontal plane on a sliding microtome. The c-Fos protein was labeled with a rabbit polyclonal

antibody raised against the amino terminus of c-Fos p62 (Santa Cruz Biotechnology, USA, sc-52) as described earlier [20, 22]. The primary antibody (1:5000) was detected by biotinylated anti-rabbit goat serum (1:1000; Jackson Laboratories, USA) and avidin-biotin complex (ABC, 1:1000; Vector Laboratories, Burlingame, CA, USA). The peroxidase reaction was developed in the presence of diaminobenzidine tetrahydrochloride (0.2 mg/ml), nickel–ammonium sulphate (0.1%) and hydrogen peroxide (0.003%) dissolved in Tris buffer.

Table 1 shows the brain structures investigated in the present study; anteroposterior levels and frame sizes are also shown. The number of investigated levels depended on the length of the particular brain region. At each level, the c-Fos signal was counted bilaterally, and the average of counts was considered. Section planes were standardized according to the atlas of Paxinos and Watson [30]. Microscopic images were digitized by an OLYMPUS CCD camera using a 10x magnification lens and stained particles were counted by means of the ImageJ v1.41o software (http://rsbweb.nih.gov/ij/). Uniform thresholds and minimum sizes of stained particles were considered.

2.4. Statistical analyses

Data are expressed as means ± SEM. C-Fos counts were square-root transformed if necessary and were analyzed by one or two-factor ANOVA as shown below. The Duncan test was used for post-hoc comparisons. Correlation between latency of muricide and neuronal activation was analyzed by using the Spearman test. Significance level was set at $p<0.05$ throughout; trends are indicated between $0.05<p<0.10$.

3. Results

3.1. Muricide

Muricide occurred suddenly without threats or other social-like signals, and was very short, almost a "point-like" behavioral act. The latency of muricidal behavior was 6.0 ± 1.2 min; its range was 1 to 10 min. 'No muricide' rats were rather passive except for occasional trials of sniffing at the mouse that showed obvious trials to avoid this contact.

3.2. Brain activation patterns

As shown above, each brain area was investigated at 2 or 3 rostro-caudal levels depending on its length. While c-Fos counts varied rostro-caudally in some areas, no interaction with group assignment was observed; therefore, the average of values obtained at these levels was considered in statistical calculi.

C-fos counts were different in the three areas of the orbitofrontal cortex ($F_{area}(2, 34)= 10.85$; $p< 0.001$). A significant group difference was also observed ($F_{group}(2, 17)= 3.34$; $p= 0.05$) but the two factors did not interact ($F_{interaction}(4, 34)< 1$; n.s.) (Fig.1a). In the medial prefrontal cortex, the three areas were also different ($F_{area}(2,38)= 68.5$; $p< 0.0001$), but no group effects were observed ($F_{group}(2, 18)= 1.72$; $p> 0.2$; $F_{interaction}(4, 36)< 1$; n.s.) (Fig. 1b). No effects were observed in the lateral septum ($F_{group}(2,18)= 2.07$; $p= 0.15$; Fig.1c).

There was a significant interaction between factors in the amygdala ($F_{area*group}(4,36) = 8.28$; $p< 0.0001$) (Fig. 2). The central and basolateral amygdala were not activated by mouse exposure, but a strong activaton was observed after muricide. The activation of the medial amygdala increased in both 'no muricide' controls and muricidal rats but the activation was stronger in the latter. By contrast, the bed nucleus of stria terminalis (BNST) was activated neither by mouse exposure nor by muricide.

A similar phenomenon was seen in the hypothalamus, where the factors interacted significantly ($F_{area*group}(4,38)= 5.46$; $p< 0.01$). C-Fos counts changed neither in the paraventricular nucleus of the hypothalmus (PVN) nor in the hypothalamic attack area

(HAA). By contrast, a strong increase in activation was observed in the lateral hypothalamus, specifically in muricidal rats (Fig. 3).

Overall, no significant group differeces were observed in the activation of the periaqueductal grey (PAG) ($F_{group}(2, 18)= 0.26$; $p=0.76$; $F_{group*area}(4, 36)= 0.73$; $p=0.57$) (Fig. 4a). However, there was an activity shift between ventral and dorsal columns at the rostral and intermediate levels, resulting in increased ventral/dorsal ratio in 'muricide' rats (rostral part: $F_{group}(2,18)=4.86$, $p<0.05$; middle part: $F_{group}(2,18)=3.34$, $p=0.059$; Fig. 4b). Monoaminergic nuclei showed no activation in either group (all regions: $F_{group}(2,18)<1.06$, ns; Table 2).

C-Fos activity in the medial amygdala, but none of the other regions, showed a positive correlation with the latency of muricide ($R=0.793$, $p<0.05$; Fig. 2).

4. Discussion

Main findings

Muricidal attacks induced strong c-Fos activation in brain areas regulating aggressive behavior which was markedly distinct from activation exhibited by rats showing no muricide during the interaction. Muricide increased the activation of the medial, central, and basolateral amygdala, and of the lateral hypothalamus. In addition, muricide shifted PAG activations from dorsal to ventral columns. This pattern of brain activation was highly congruent with findings obtained in cat stimulation studies, was in several respects similar to c-Fos findings obtained with the rat cockroach-hunting model, and was highly similar with the findings obtained in rats submitted to the glucocorticoid dysfunction-model of abnormal aggression (Fig. 5). In the rat models, overactivations were seen in brain areas that promoted predatory attacks in cats. In brain areas where electrical stimulation inhibited predatory aggression in cats, no activation was seen in any of the rat models, except for the medial amygdala, which was activated by muricide. However, this brain response correlated positively with the latency

of muricide, suggesting that similar to cats, the medial amygdala is involved in the negative modulation of predatory aggression. Taken together, our findings show that brain mechanisms underlying predatory aggression are highly conservative, and strongly support our earlier suggestions on the similarities between the brain mechanisms of predatory aggression and those underlying certain forms of abnormal social aggression.

Comparisons with earlier findings

The immediate-early gene product c-Fos has been widely used as an indicator of acute neuronal activation to describe brain activation patterns during different aggressive interactions in several laboratory species, including mice, hamsters and rats [31-34]. It was also used to study the brain mechanisms of cockroach hunting, another model of rat predatory aggression [27, 28]. The study presented here was performed to serve as a comparison for other models of predatory aggression (stimulation-induced aggression in cats [1, 4], and cockroach hunting in rats [27, 28]), as well as for certain models of abnormal aggression in rats (glucocorticoid dysfunction model, [18, 21, 35]). These models will be compared by starting with the hypothalamus that plays crucial roles in the elicitation of biting attacks in both cats and rats [1].

Cholinergic drugs injected into the lateral hypothalamus elicited muricide in rats [23, 26], whereas the electrical stimulation of this brain area induced quiet biting in cats, a predatory form of aggression in this species [1, 6, 24, 36, 37]. In our muricidal rats, strong and highly muricide-specific activation was seen in the lateral hypothalamus along its whole rostro-caudal extent, which supports the notion that this region is a primary center for predatory aggression. In the cockroach-hunting model, this region was also activated; however, its activity was not specific to hunting as the effects of feeding (the control treatment in this model) were similarly strong [28]. Indeed, the lateral hypothalamus is crucially involved in feeding [38]; hence, one can assume that the effects of cockroach killing

were masked by the effects of food intake. Interestingly, the lateral hypothalamus was also overactivated in the glucocorticoid dysfunction model of abnormal aggression, the subjects of which deliver bites to highly vulnerable targets of their opponents (head, throat and belly) on the background of diminished social signaling of attacks by threats, which make their behavior similar to that seen during predation [22]. In contrast to the lateral hypothalamic region, the mediobasal hypothalamus (often called the 'hypothalamic attack area' in rats) elicits affective aggression in both cats and rats, and is activated by resident/intruder interactions in the latter species [1, 4, 21, 37, 39, 40]. Cat studies revealed a reciprocal inhibition between the lateral and mediobasal regions, which involves a context-dependent control of attacks, making social and predatory aggressions mutually exclusive [4, 39]. Although similar direct evidence is missing in rats, one cannot rule out a potentially similar inhibitory mechanism; for instance, mediobasal hypothalamic lesions result in ’uncontrolled’ muricidal behavior which suggests cross-species similarities [41]. The mediobasal hypothalamic area was activated neither in our muricidal rats, nor in the cockroach-hunting model [28]. In the glucocorticoid dysfunction model, the mediobasal hypothalamus was activated by fights as compared to non-fighting controls, but its activation was similar to that seen in sham operated fighting controls. Taken together, these findings suggest that the mediobasal hypothalamus plays no particular roles in muricide, cockroach hunting, or in the abnormal attack features of rats submitted to the glucocorticoid-dysfunction model. The activation of the paraventricular nucleus of the hypothalamus –a brain region that orchestrates the glucocorticoid stress response– was activated in neither rat model discussed here, which is in line with the low emotionality of both predatory aggression and glucocorticoid dysfunction-induced abnormal aggression [18, 19, 28].

Hypothalamic attack areas receive inputs from the amygdala, which plays a subregion-specific role in aggression control. In cats, the medial amygdala and its major output region

BNST inhibit predatory attacks, whereas the central and basolateral nuclei facilitate this response [1, 42-46]. Our findings are in line with these reports by showing a muricide-specific activation of the central and basolateral nuclei. Highy similar findings were obtained in the cockroach-hunting model of predatory aggression [28]. The over-activation of the central nucleus is also well-documented in the glucocorticoid-dysfunction model, where predatory-like attacks are shown in a social context [19, 21, 47]. No medial amygdala activation surpassing that seen in controls were observed in the rat models discussed here except for muricide. Our correlation analysis, however, suggests that this region is involved in the negative modulation of muricide, which is congruent with findings obtained in cats, where the concommitant stimulation of the medial amygdala and lateral hypothalamus decreased the efficacy of the latter in inducing predatory attacks [1].

Hypothalamic attack centers send projections to the PAG, which controls behaviors and autonomic functions during aggressive conflicts [1]. This brain area is organized in columns, which are implicated in behavioral control diferentially. In cats, the stimulation of ventral columns (lateral and ventrolateral PAG) induces predatory attacks, whereas dorsal columns (dorsomedial and dorsolateral PAG) are involved in defensive responses [1, 6, 48-50]. Here we found that muricidal attacks were accompanied by a dominantly ventral activation of the PAG. Congruent findings were obtained in the cockroach-hunting model [27, 28, 51]. A comparison of cockroach hunting- and muricide-induced c-Fos activity also suggests a rostro-caudal heterogeneity of the PAG and a potential role of rostral regions in the switch between intraspecific and predatory attack patterns. Noteworthy, a ventral shift in PAG activation was also seen in the abnormal aggression model [21, 22, 35, 47, 52].

Our findings are equivocal as it regards the involvement of prefrontal regions in aggression control. Muricide increased the activation of the orbitofrontal cortex as compared to 'no-mouse' controls but not as compared to 'no-muricide' controls. A similar situation

occurred in the glucocorticoid dysfunction model, where aggression did increase the activation of the orbitofrontal cortex as compared to non-fighting controls, but this activation was not larger than that seen in sham operated fighting controls [35]. The orbitofrontal cortex was not investigated in the cockroach model so far, but findings on the activation of the medial prefrontal cortex were similarly equivocal. The activation of this area was larger in cockroach-hunting rats as compared with home cage controls, but smaller than that seen in feeding controls [28]. These findings suggest that the involvement of the prefrontal cortex in aggression is more complex then expected, and invites further research.

Conclusions

(i) The neural underpinnings of predatory aggression show remarkable cross-species similarities. Muricide enhanced the activation of brain areas which promote, but did not affect the activation of areas that inhibit predatory aggression in cats (Fig. 5). Although enhanced activation was seen in the medial amygdala, which inhibits predatory attacks in cats, this brain region appears to modulate muricide negatively as shown by correlation analysis.

(ii) The pray has an impact on brain activation patterns. The effects of cockroach hunting and muricide were similar in some, but not in all brain regions. As the food intake component of cockroach hunting was controlled in the study by Comoli et al. [28], differences in brain activations are likely due to behavioral differences inherently associated with the nature of the pray. According to our observations, explicit acts of killing are present in the muricide, but mostly absent in cockroach hunting model. Still, the brain mechanisms underlying the two forms of rat predation overlap to a certain extent showing that different forms of predation are controlled by overlapping but not entirely similar networks.

(iii) Brain activation patterns elicited by muricide and glucocorticoid dysfunction-induced abnormal aggression were highly similar, and support the notion that this type of abnormal aggression activates brain mechanisms involved in predation. This parallels

behavioral and physiological similarities; in both models, attacks are targetted on vulnerable body parts, social communication is deficient, and physiological arousal is low. These observations support earlier clinical descriptions that differentiate reactive/affective from *predatory*/instrumental types of human aggression, and implicate that their neural underpinnings are different [7-9].

The present study used a 'descriptive mapping' approach. Future studies are required to elucidate the causality between brain activation patterns and aggressive behavior.

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Table 1. The investigated brain areas.

Area	Level of analysis (mm from Bregma)	Number of slices	Frame size (mm^2)
Orbitofrontal cortex	4.20 to 3.70	2	0.564
Medial prefrontal cortex	3.20 to 2.70	2	0.469
Lateral septum	0.70 to 0.20	3	0.880
Mediobasal hypothalamus ("hypothalamic attack area")	-1.60 to -2.30	3	0.575
Lateral hypothalamic area	-1.88 to -3.60	3	0.673
Paraventricular nucleus of the hypothalamus (parvocellular region)	-1.40 to -1.80	3	0.161
Medial amygdala	-2.30 to -2.80	2	0.697
Central amygdala	-1.80 to -2.56	3	0.576
Basolateral amygdala	-2.30 to -3.14	3	0.748
Bed nucleus of the stria terminalis, medial part	-0.80 to -0.92	2	0.403
Ventral tegmental area	-5.20 to -6.04	2	0.733
Dorsal raphe	-7.64 to -8.00	2	0.368
Locus coeruleus	-9.80 to -10.04	2	0.149
Periaqeductal grey, dorsomedial part	-6.72 to -8.30	3	0.184
Periaqeductal grey, dorsolateral part	-6.72 to -7.80	2	0.207
Periaqeductal grey, lateral part	-6.72 to -8.30	3	0.282
Periaqeductal grey, ventrolateral part	-7.80 to -8.30	2	0.368

Note that the relatively broad spatial range of section planes is explained by the fact that 2-3 sections were investigated in the case of each area, to invetigate potential rostro-caudal differences in activation patterns. Post-hoc analysis showed no such differences; therefore, c-Fos counts were averaged over levels. The periaqueductal gray was an exception in this respect, because the structure of this brain area changes substantially in the rostro-caudal direction. Section planes were standardized according to the Rat Brain Atlas of Paxinos and Watson [30]

Table 2. Muricide-related neuronal activity in monoaminergic nuclei as indicated by c-Fos expression.

Area	No mouse	No muricide	Muricide
Dorsal raphe	18.79±5.20	19.43±4.61	22.19±3.64
Locus coeruleus	4.93±0.91	4.83±1.06	5.91±1.17
Ventral tegmental area	16.79±4.39	15.36±3.28	21.44±6.32

Data (mean ± standard error of the mean) shows c-Fos counts in the frames specified in Table 1. No statistically significant differences were observed.

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Figure captions and legends

Fig. 1. Neuronal activity in the (a) orbitofrontal and (b) medial prefrontal cortex as well as (c) the lateral septum as indicated by c-Fos expression. The schematics illustrate the regions investigated (gray squares) based on Paxinos and Watson [30]. *ac*, anterior commisure; *cc*, corpus callosum; *Cg1*, anterior cingulate cortex; *fmi*, forceps minor of corpus callosum; *IL*, infralimbic cortex; *LS*, lateral septum*; MO*, medial orbitofrontal cortex; *mPFC*, medial prefronal cortex; *LO*, lateral orbitofrontal cortex; *OB*, olfactory bulb; *PrL*, prelimbic cortex; *VO*, ventral orbitofrontal cortex; *, significant difference from 'no mouse' controls ($p < 0.05$).

Fig. 2. Neuronal activity in the extended amygdala. The schematics illustrate the regions investigated (gray ovals) based on Paxinos and Watson [30]. Panel 'b' shows correlation between medial amygdala activity and the latency of muricide. *MeA*, medial amygdala. *ac*, anterior commisure; *BLA*, basolateral amygdala; *BNST*, bed nucleus of stria terminalis, medial part; *CeA*, central amygdala; *ic*, internal capsule; *MeA*, medial amygdala; *ot*, optic tract; *st*, stria terminalis; *, significant difference from 'no mouse' controls; $^{\#}$, significant difference from 'no muricide' controls ($p < 0.05$).

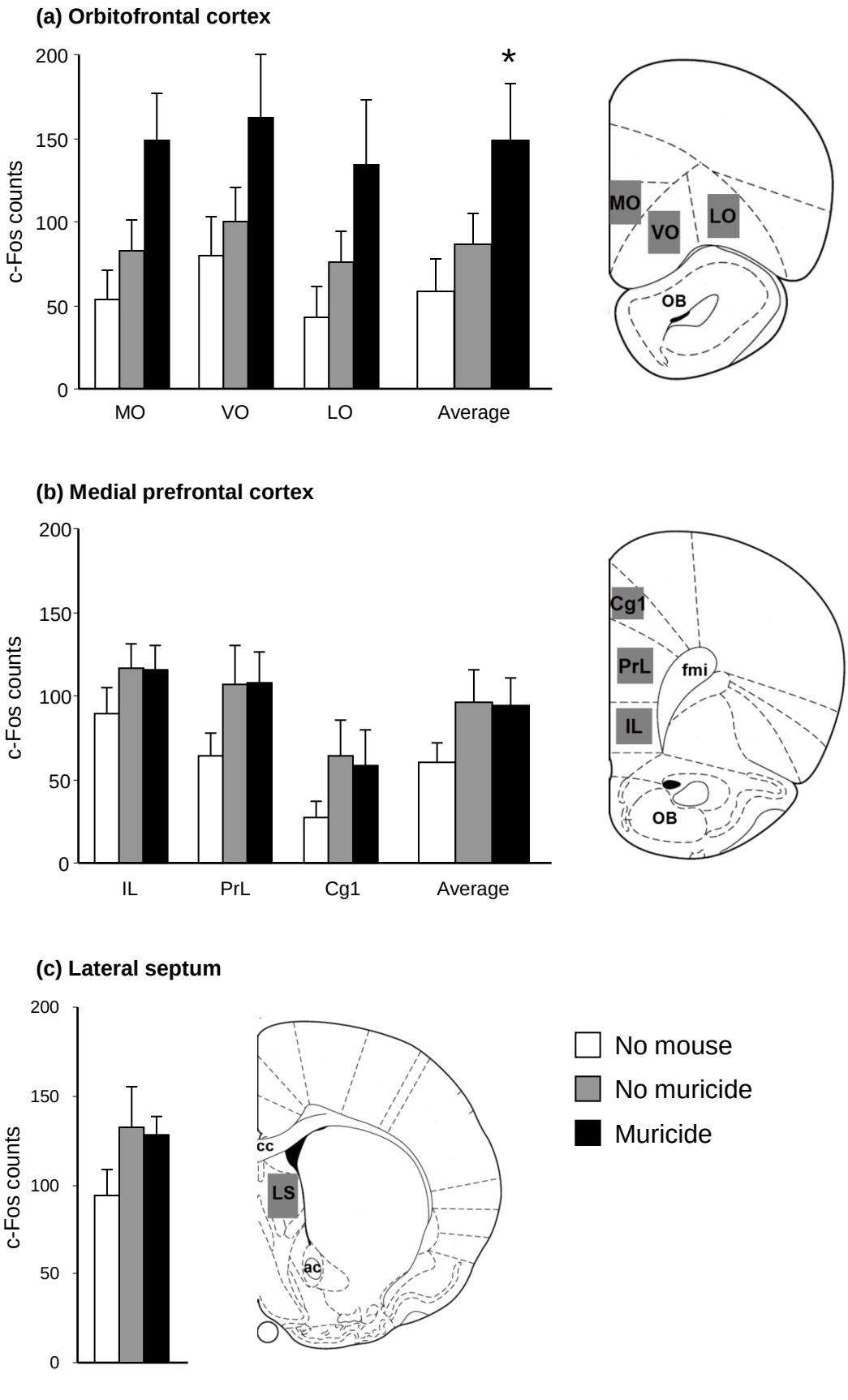
Fig. 3. Hypothalamic neuronal activity following an interaction with a mouse as indicated by c-Fos expression. The schematics illustrate the regions investigated (gray ovals) based on Paxinos and Watson [30]. *f*, fornix; *HAA*, hypothalamic attack area (mediobasal hypothalamus); *LH*, lateral hypothalamus; *ot*, optic tract; *PVN*, paraventricular nucleus of the hypothalamus; *, significant difference from 'no mouse' controls; $^{\#}$, significant difference from 'no muricide' controls ($p < 0.05$).

Fig. 4. Overall c-Fos counts (a) and the ventral/dorsal ratio of activation (b) at different rostro-caudal levels of the periaqeductal grey. The schematics illustrate the regions investigated (gray ovals) based on Paxinos and Watson [30]. *DL*, dorsolateral part; *DM*, dorsomedial part; *L*, lateral part; *VL*, ventrolateral part; *V/D*; ventral/dorsal ratio of activation; *, significant difference from 'no mouse' controls ($p < 0.05$); $^{+}$, marginally significant difference from 'no mouse' controls ($0.1 > p < 0.05$); $^{\#}$, significant difference from 'no muricide' controls ($p < 0.05$).

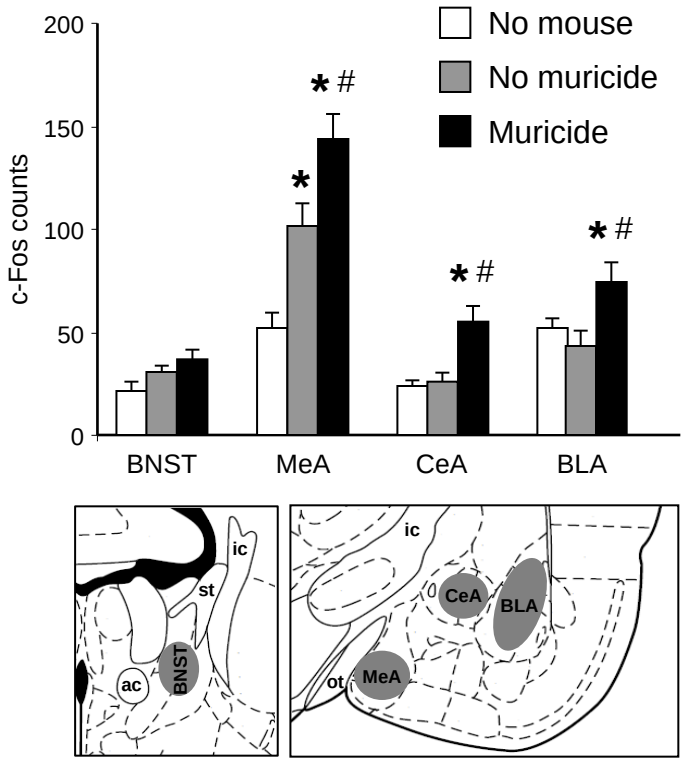
Fig. 5. A comparison of four aggression models. Data obtained in the muricide (based on present findings), cockroach hunting (based on [20, 21]), and glucocorticoid dysfunction induced abnormal aggression models (based on [11, 14, 52]) were grouped according to the effects of electrical stimulation on predatory aggression in cats (based on [1, 2]). *OFC*, orbitofrontal cortex; *mPFC*, medial prefrontal cortex; *LS*, lateral septum; *BNST*, bed nucleus striae terminalis; *HAA*, hypothalamic attack area (mediobasal hypothalamus); *MeA*, medial amygdala; *CeA*, central amygdala; *BLA*, basolateral amygdala; *LH*, lateral hypothalamus; *PAG*, periaqueductal gray; *horizontal gray arrows*, no change in c-Fos activation; *vertical black arrows*, increased c-Fos activation; * *and* ◦, similar and discrepant, respectively, findings in muricide and cockroach-hunting models; *empty cells*, no data.

Note. The following contrasts were considered: no muricide vs. muricide (mouse killing); feeding vs. cockroach hunting (cockroach hunting); sham-operated fighting vs. adrenalecomized fighting (glucocorticoid dysfunction-induced abnormal aggression).

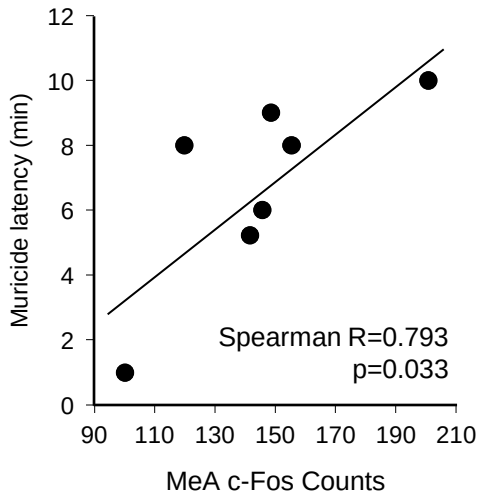
Figure(s)

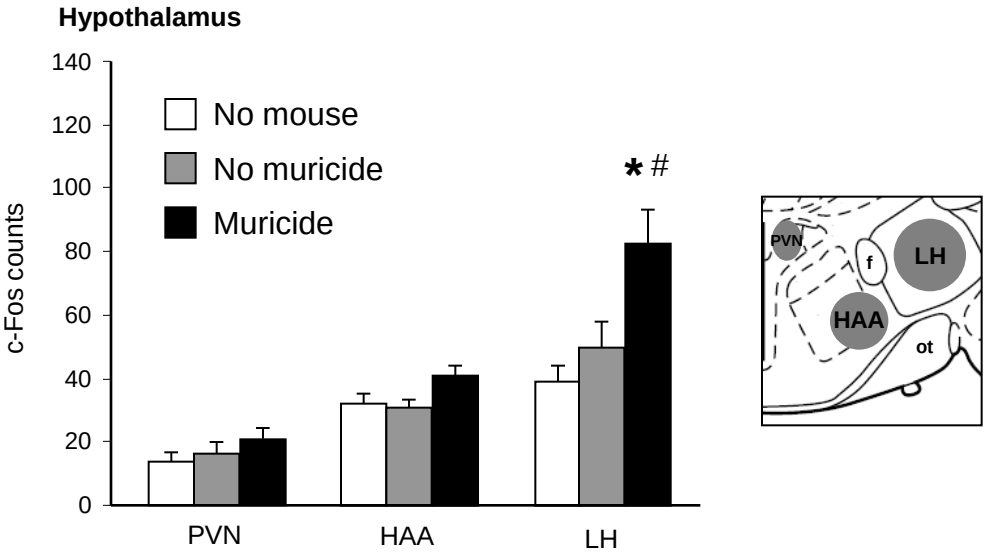

(a) Orbitofrontal cortex
200
150
100
50
0
c-Fos counts
*
MO
VO
LO
Average
MO
VO
LO
OB
(b) Medial prefrontal cortex
200
150
100
50
0
c-Fos counts
IL
PrL
Cg1
Average
Cg1
PrL
fmi
IL
OB
(c) Lateral septum
200
150
100
50
0
c-Fos counts
cc
LS
ac
No mouse
No muricide
Muricide

(a) Extended amygdala

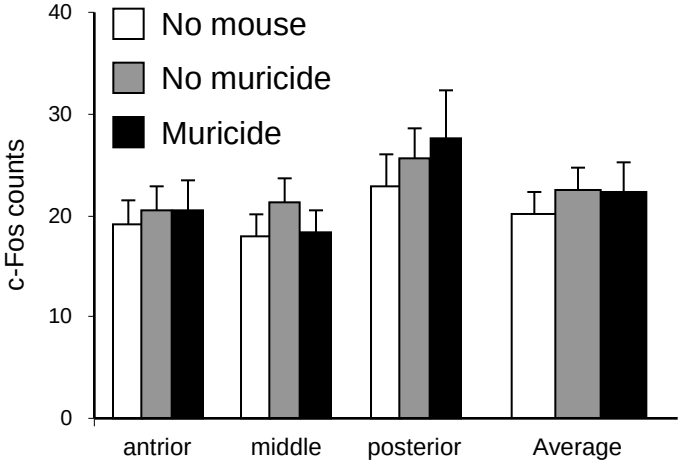


(b) Correlation between MeA activation and muricide

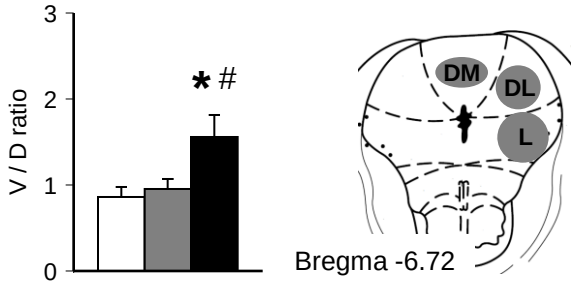


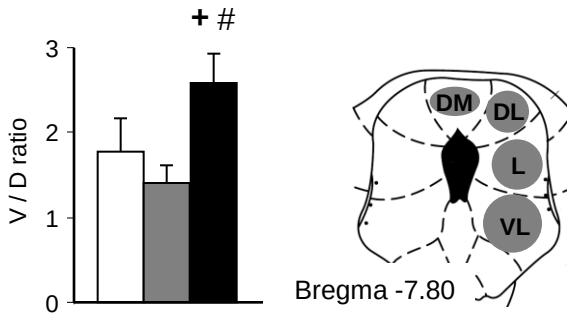

Hypothalamus
c-Fos counts
140
120
100
80
60
40
20
0
No mouse
No muricide
Muricide
* #
PVN
HAA
LH
PVN
f
LH
HAA
ot

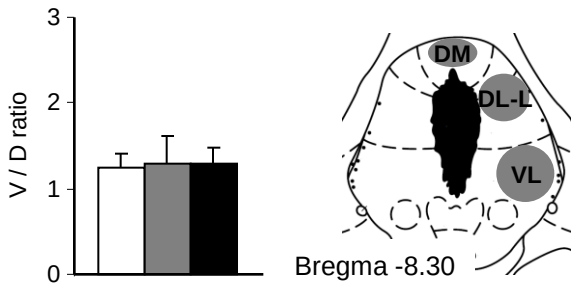
(a) Periaqueductal gray – activation levels



(b) Periaqueductal gray – ventral/dorsal activation ratio







A comparison of the models considered here

Areas that *inhibit* predatory attacks in cats	**Mouse killing Rat**	**Abnormal aggression Rat**
OFC	→	→
mPFC	→ °	→
LS	→ *	→
BNST	→ °	→
HAA	→ *	→
MeA	↑ °	→
Areas that *stimulate* predatory attacks in cats		
CeA	↑ *	↑
BLA	↑ *	
LH	↑ °	↑
ventral PAG	ventral shift in PAG activation *	ventral shift in PAG activation