

INCREASED SWEET TASTE PREFERENCE FOLLOWING THE LESION OF BASOLATERAL NUCLEUS OF AMYGDALA (BLA) IN RAT

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Abstract : A study of gustatory preference was carried out in Wistar strain albino rats by electrolytically lesioning the basolateral nucleus of amygdala. The intake of sweet tasting saccharin (Sn-0.1% soln w/v), NaCl solutions (1% Soln w/v) and tap water, were tested in single bottle, 2 bottle choice and 3 bottle choice situations. The consumption of fluids both before and after the stereotaxic surgery was recorded and statistically analysed. Lesion of BLA increased the intake of all fluids in the single bottle tests ($P < 0.01$). But the increase in the group provided with saccharin was more than that in other two groups. When taste preference was tested using 2-bottle and 3-bottle choice situations, the lesioned rats were seeking Sn solution. In our previous study, we found that the lesion of BLA increased fluid intake in rats. The intake, in the present study was more in those animals provided with Sn solution. When choice was given, the rats shifted their preference from water to the Sn following the lesion. Thus this study confirms that BLA is involved in the preference for sweet tasting solution over the NaCl or plain water.

Key words : amygdala
lesion

basolateral nucleus
taste preference

INTRODUCTION

The forebrain structures implicated in gustatory mechanism included the nuclear groups of amygdala, namely basolateral (1), central (2) and also the structures in the hypothalamus - the lateral hypothalamus and the ventromedial nucleus (3). In the brainstem, structures such as nucleus of tractus solitarius (NTS) (4), parabrachial nucleus (PBN) were also considered to be important loci involved in the gustatory

responses (5). The central gustatory afferent system could provide an anatomical framework that links most of the forebrain areas which were implicated in the control of food and water intake, and ingestion or rejection behaviours with that of brain stem areas (5). The neural mechanisms underlying feeding, drinking and pleasure were located in the hypothalamus (6,7). Apart from the cortical connections, the identification of a parallel gustatory pathway which projected to the ventral

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forebrain from the PBN, ramifying to the amygdala and substantia innominata, complicated the understanding of the central gustatory pathways (5).

In this context, the role of amygdaloid nuclei in the taste perception responses attain significance, but the reports available are highly contradictory. Electrolytic lesions made in the central nucleus of amygdala reportedly decreased the preference to sweet solution in rats (2, 8), while Kemble and associates (9) reported no significant variations in the intake of taste solution following complete amygdalaectomy. It was reported that lesion of lateral amygdala alone produced an increased intake of sweetened solutions (10), whereas, Siegel et al. (1) reported a decreased sucrose solution consumption following ibotenic acid induced lesion of basolateral amygdala.

The available literature on this subject appears vague and seemingly nonspecific areas of amygdaloid nucleus were investigated for the involvement in taste related activities. In the present study, restricted lesions of the basolateral nucleus of amygdala was performed. The intake of sweet solution (0.1% of saccharin, w/v) and salty solution (1% NaCl, w/v) and tap water, were studied before and after the lesion, by providing single, two and three bottle choice experiments. The three bottle choice was unique and hence the specific role of the basolateral nucleus of amygdala could be elucidated with particular reference to the taste preference.

METHODS

The Wistar strain male albino rats, weighing about 210-230 gms were used for

this study. These rats were housed in individual metallic cages under standard laboratory conditions. Food pellets and fluid in drinking bottles were made available *ad lib*. These animals were divided into three experimental groups as follows :

1. Single bottle Controls/Experimental (n=60) : Sn=20; NaCl=20; Water=20.
2. 2-Bottle choice test : (n=40) : Sn + Water = 20; NaCl + Water = 20.
3. 3-Bottle choice test = (n=22).

In each group, half of the total number of animals were maintained as sham lesioned (SL) control group.

The lesion of the BLA was performed using the Horsely-Clarke stereotaxic apparatus, according to the stereotaxic co-ordinates for rat brain (11).

Stereotaxic co-ordinates for BLA were AP = -3.14 mm behind the bregma; L = \pm 4.8 mm from the midline; V = 8.5 mm from the surface of the skull. Burr holes were made in the skull after anaesthetising the animal using Nembutal (40 mg/kg body weight ip). The stereotaxic lesion was carried out by passing a DC current of 2mA for 20 sec using a Grass (USA) lesion maker. The stainless steel electrode (22 gauge steel wire) was insulated except for the 0.5 mm at the tip. The cathode was connected to the tail of the rat. The sham operated control animals underwent the surgical procedure only and no current was passed. Following the lesion, two days were allowed for recuperation. The data from those animals with histologically confirmed

bilateral lesions only were accepted for the analysis of the results. Diagrammatic reconstruction of the serial sections of the brain were made to show the extent of the lesion (Fig. 1).

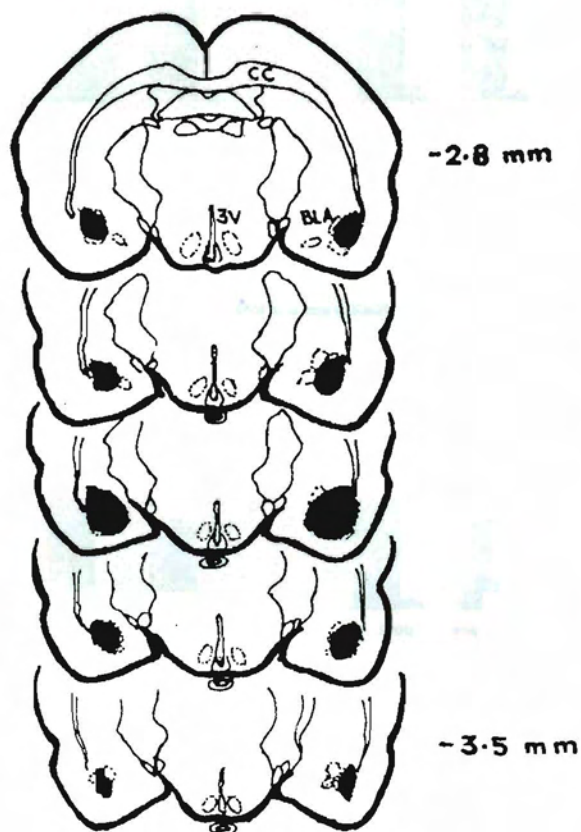


Fig. 1: Serial sections of the rat brain showing the site of lesion at the BLA.

CC-corpora collosum; 3v-3rd ventricle

Measurements

The rats were provided with measured quantity of water, NaCl or saccharin (Sn) in the single drinking bottle. Daily intake of the fluid for each animal in the respective groups, was recorded at 10.00 AM everyday for 7 days before the lesion and for 21 days

(divided into 3 weeks) following the electrolytic lesion.

In the two bottle and the three bottle choice tests, the measurements of individual fluids were recorded in the prelesion period (7 days) as well as postlesion period (15 days). The total fluid intake (in all the bottles provided to a rat) was calculated and recorded in the respective periods.

Statistical analysis :

The non-parametric statistics for behavioural studies was applied to assess the statistical significance of the data.

1. Comparison of prelesion with that of postlesion data was done by applying the Wilcoxon 2-tailed signed rank sum test. ($P < 0.01$ was considered as significant).
2. Intergroup comparison was done by applying Mann-Whitney 'U' test. ($P < 0.001$ was considered as significant).

RESULTS

After BLA lesion a significant increase in the fluid intake of the rats in all groups, viz. Sn group, NaCl group as well as the water group (Table I, Wilcoxon test, $P < 0.01$) was observed. In the group provided with NaCl, the daily intake in the prelesion period was higher than the other two groups. The rats in water group and NaCl group consumed as such as 30% more than the prelesion levels. The increase in consumption of saccharin group was more than that in the other two groups (Table I). Intergroup comparison of fluid

TABLE I: Intake of Sn, NaCl and water in single bottle test. pre = prelesion; post = postlesion. Values in ml \pm SE. ** - $P < 0.01$ (Wilcoxon test) compared to the prelesion levels.

Fluids ml/day	Control		Experimental	
	Pre	Post	Pre	Post
Sn	19.0 \pm 0.7	19.5 \pm 0.6	18.2 \pm 0.6	25.5 \pm 1.5**
NaCl	23.2 \pm 0.6	23.3 \pm 0.6	24.1 \pm 0.4	26.8 \pm 0.4**
Water	19.3 \pm 0.5	19.5 \pm 0.5	18.0 \pm 0.4	22.5 \pm 0.3**

consumption with that of SL controls showed a significant variation (Mann-Whitney 'U' Test $P < 0.001$).

In the two bottle choice test, the rats provided with a choice of Sn and Water (Sn+Water group) in two different drinking bottles consumed significantly more saccharin than water after the lesion ($P < 0.01$, Fig. 2). Their water consumption got reduced dramatically following the lesion. However, the total fluid intake in such lesioned rats were significantly higher than that in the prelesion period ($P < 0.01$ compared to prelesion and $P < 0.001$ when compared to the SL control group). 'NaCl+Water' group of rats consumed only water before and after the lesion. Water consumption was significantly more following the lesion of BLA ($P < 0.01$, Wilcoxon test, Fig. 3). They did not consume NaCl solution at all.

When all the three solutions (Sn, NaCl and water) were made available simultaneously (in 3 bottles), the rats consumed only water in the prelesion period, and in the SL control rats. However, the rats lesioned in BLA consumed saccharin solution in significantly larger

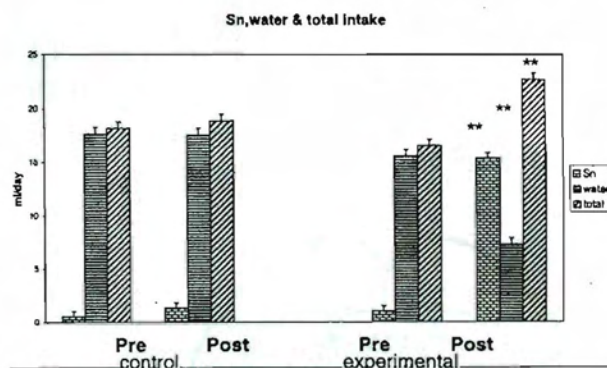


Fig. 2: Intake of Sn & water in two bottle choice test. pre = prelesion; post = postlesion. ** - $P < 0.01$ compared to the prelesion levels.

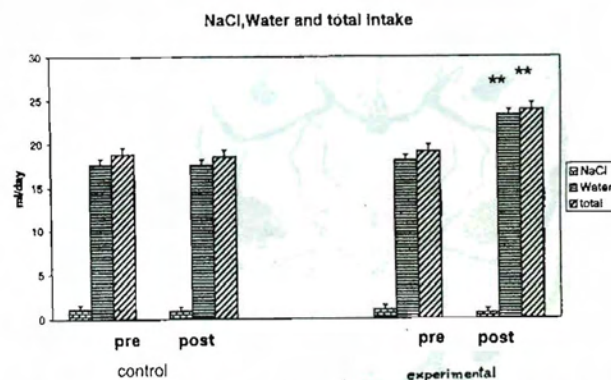


Fig. 3: Intake of NaCl & water in two bottle choice test. pre = prelesion; post = postlesion. ** - $P < 0.01$ compared to the prelesion levels.

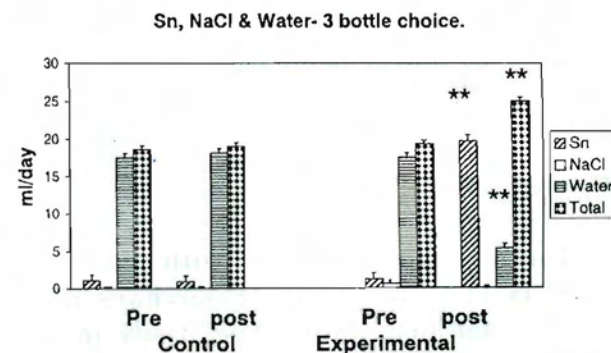


Fig. 4: Intake of Sn, NaCl & water in three bottle choice test. pre = prelesion; post = postlesion. ** - $P < 0.01$ compared to the prelesion levels.

volumes and their intake of water got reduced ($P < 0.01$, Fig. 4). The total fluid consumption was however more following the lesion ($P < 0.01$).

DISCUSSION

From the results of our experiments, it appears that the basolateral nucleus of amygdala is one of the principal centres in the amygdaloid complex, which may be involved in the selection of sweet tasting substances. Rolls and Rolls (10) have demonstrated that the rats lesioned in large areas of the amygdala showed a taste preference towards saccharin solution. This was also later confirmed by others (12). It has also been demonstrated that the learned taste aversion acquired before the lesion of basolateral amygdaloid nucleus was not retained following the lesion (12). The observations of Black and Weingarten (13) revealed that the disruption of the stria terminalis (ST) could produce dramatic changes in the taste reactivity responses in rats. ST is an important efferent pathway from amygdala to the hypothalamus. In our present study, the rats consumed more fluid after the lesion of basolateral nucleus of amygdala, when the rats were presented with Sn, NaCl or Water in single bottle, two bottle and three bottles. This observation was in agreement with our earlier report (14). But there was striking increase in the fluid consumption in the Sn group, which was considerably more than that in the other two groups (Table I).

The observation of increased intake of saccharin in excess of the other two types of fluids, prompted us to carry out the two-bottle choice test, to detect whether the preference of the rats got altered as a result

of the lesion of basolateral amygdala. While the rats consumed only water in the prelesion period, the BLA lesioned rats shifted their preference to Sn solution in the Sn-Water group. The Sham lesioned control rats in Sn-Water group continued to consume water throughout the duration of the experiment (Fig. 2). From the results of our experiments, it appeared that the alterations of taste preference was restricted to the Sn intake but not to salty taste, because the rats belonging to NaCl-Water group consumed only water all along the duration of the experiment. Thus, the present finding clearly indicated that the lesion of the BLA not only increased the intake of fluids but also that such lesioned rats preferred to consume sweet tasting solution against water and NaCl.

Further it was decided to carry out the 3-bottle choice test, providing Sn, NaCl and Water in 3 different drinking bottles simultaneously. The 3-bottles free choice situation was considered to be a tougher task to the small animals such as rats. Thus, this test would assess the intensity of their urge to seek sweet tasting solution in the presence of other solutions as well. The positions of the bottles were changed randomly everyday. In the 3-bottle choice test also, the rats, which were consuming only water before lesion of BLA, shifted their preference to saccharin solution (Fig. 3). This provided us the conclusive evidence of increased sweet taste preference in the BLA lesioned rats. Further work is being carried out with lesioning of central group of neurons of the amygdala and possibly stria terminalis of the limbic system to prove unequivocally the role of specific nuclei of amygdala in determining the taste preference.

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