**Changes in Electroencephalogram of the Rat Following Olfactory Bullectomy**

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WATANABE, S., FUKUDA, T. and UEKI, S. Changes in Electroencephalogram of the Rat Following Olfactory Bullectomy. Tohoku J. exp. Med., 1980, 130 (1), 41-48 —— The changes in electrical activities following olfactory bullectomy were investigated in rats with chronically implanted electrodes, in comparison with those induced by septal lesions. Electroencephalogram (EEG) of the amygdala changed to low voltage fast waves immediately after bilateral olfactory bullectomy, while EEG of the neocortex gradually showed an arousal pattern consisted of low voltage fast waves, and hippocampal theta waves were markedly synchronized during the period of a week or so after bullectomy accompanied with the appearance of characteristic hyperemotionality. No change was observed in EEG of the septum, hypothalamus or the midbrain reticular formation. Following unilateral olfactory bullectomy, no behavioral changes occurred, but EEG of the ipsilateral amygdala showed low voltage fast activity without causing any EEG changes in other brain structures. Bilateral septal lesions immediately caused hyperreactivity in rats which gradually disappeared during the course of a week or two after the surgery. The hippocampal theta activity completely disappeared and EEG turned to low voltage fast waves immediately after septal lesions, but no significant EEG change was observed in the amygdala or the neocortex. This change in hippocampal EEG continued for a long time even after hyperreactivity disappeared. —— electroencephalogram; olfactory bullectomy; amygdala; septal lesion; hyperemotionality

It has been well documented that bilateral olfactory bullectomy induces characteristic hyperemotionality or aggressiveness in rats (Watson 1907; Vergnes and Karli 1963; Myer 1964; Ueki and Sugano 1965; Ueki et al. 1972a). This hyperemotionality has also been used for evaluating the effects of psychotropic drugs (Kumadaki et al. 1967; Ueki et al. 1972b). However, the neural mechanisms underlying this hyperemotionality are still unknown, although Douglas et al. (1969) have postulated that the activity change in the amygdala is important, while Nurimoto et al. (1974) have emphasized the importance of the olfactory tubercle in the development of hyperemotionality after bilateral olfactory bullectomy.

No study has been so far done concerning the electroencephalographic (EEG) changes induced by olfactory bullectomy. In the present study, the authors

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aimed to investigate the changes in EEG of various brain areas, accompanied with the occurrence of hyperemotionality, following olfactory bulbectomy in the rats with chronic electrode implants, and compared with those induced by septal lesions, in the hope of elucidating the neural mechanism of hyperemotionality.

**METHOD**

A total of 34 adult male Wistar-King A rats, supplied by Kyushu University Institute of Experimental Animal, weighing 250-300 g at the beginning of the experiment, were used.

The animals were anesthetized with sodium pentobarbital 40 mg/kg i.p. and bipolar electrodes made of stainless steel wire of 0.2 mm in diameter were first implanted chronically in several brain structures, according to the brain atlas of König and Klippel (1963). The stereotaxic coordinate was A 7.5, L 0.5, V +1.0 for the septum; A 5.0—5.5, L 4.0, V −3.0—−4.0 for the amygdala; A 2.0—−2.5, L 4.0—5.0, V +2.0—−2.5 for the hippocampus; A 4.0, L 0.5, V −3.5—−4.0 for the hypothalamus, and A 2.0, L 2.0, V −1.5 for the midbrain reticular formation. Cortical electrodes were placed on the frontal cortex.

Olfactory bulb ablations and septal lesions were performed under pentobarbital anesthesia (40 mg/kg i.p.). The olfactory bulb was removed surgically by suctioning after trepanation of the skull just above the bulb. The septum was lesioned by electrocoagulation with DC current of 2 mA applied for 20 sec through the implanted electrodes. Penicillin (100,000 units/animal) was given intramuscularly for 3 days after the surgery.

EEG was recorded on the polygraph (model 602, Sanei Sokki) at the condition of freely moving in a shielded cage with simultaneous observation of behavior of the rat.

The position of electrodes and the extent of brain lesions were histologically verified after the experiments were terminated.

Recording of spontaneous EEG in various brain areas was repeated for about 1 month before brain lesioning and continued for approximately 3 months after surgery, simultaneously with the observation of behavioral changes of the animals.

**RESULTS**

*EEG changes following bilateral olfactory bulbectomy*

Seventeen male rats were used in this experiment, and the spontaneous EEG was compared before and after bilateral olfactory bulbectomy. When intact rats were placed in the shielded cage for EEG recording, they moved around the cage showing exploratory behavior. In this period, EEG of the neocortex and amygdala showed an arousal pattern characterized by low voltage (50–100 μV) fast waves (10–20 Hz), and spikes and spindle bursts were often observed in the amygdala (Fig. 1). Hippocampal EEG showed well synchronized theta waves of high voltage (100–200 μV). EEG of the septum, hypothalamus and midbrain reticular formation showed low voltage fast waves. Within a period of 10–30 min after EEG recording was started in a shield cage, locomotion of the rat gradually decreased. In this period, high voltage fast waves increased in the neocortical EEG and the hippocampal theta rhythm was desynchronized. After EEG recording was repeated several times in the period of about 1 month, the olfactory bulbs were bilaterally removed. Within a few days after bilateral olfactory bulbectomy no behavioral changes were observed in most of the rats, but locomotor activity and exploratory behavior such as sniffing and rearing gradually increased during the
EEG of Olfactory Bullectomized Rats

Fig. 1. Effect of bilateral olfactory bulbectomy on EEG activity in the rat.
A: control EEG. B, C and D: 1 day, 5 days and 15 days, respectively, after bilateral olfactory bulbectomy.
AM-R, right amygdala; AM-L, left amygdala; CO, frontal cortex.
Abbreviations are the same for all figures.

Course of 1 week after bulbectomy. Furthermore, aggressive behavior also developed; i.e. the rats attacked a rod presented in front of the nostril and some of them showed muricide. However, hyperreactivity such as jumping, flight and squeak responses to given stimuli was less marked. In accordance with the appearance of hyperactivity and aggressiveness, the neocortical EEG showed much faster waves of low voltage and the hippocampal theta rhythm was more markedly synchronized as compared with those seen before olfactory bulbectomy. These EEG changes remained throughout the observation period of 1 month.

On the other hand, in EEG of the amygdala, the voltage was significantly decreased immediately after bilateral olfactory removal in all animals and spikes and spindle bursts were slightly increased in 10 out of 17 rats. After about 1 week, EEG of the amygdala gradually recovered its voltage in about a half of the animals operated, while low EEG voltage remained throughout the experiment in the rest of the animals (Fig. 2). However, no difference in behavioral changes was found between these 2 groups. No significant EEG changes were observed in the septum, hypothalamus or the midbrain reticular formation after bilateral olfactory bulbectomy.

EEG changes after unilateral olfactory bulbectomy

Changes in EEG after unilateral ablations of the olfactory bulb were examined in 8 rats in which chronic electrodes were implanted either in the neocortex and amygdala of both sides or in the neocortex and hippocampus of both sides. The rats showed no significant behavioral changes after unilateral olfactory bulbectomy, differently from those with bilateral bulbectomy. No EEG change was observed in the neocortex or the hippocampus, but EEG in the amygdala
Fig. 2. Effect of bilateral olfactory bulbectomy on EEG activity in the rat. A: control EEG. B, C and D: 5 days, 11 days and 15 days, respectively, after bilateral olfactory bulbectomy. RF, midbrain reticular formation; HC, hippocampus.

Fig. 3. Effect of unilateral olfactory bulbectomy on EEG activity in the rat. A: control EEG. B, C and D: 6 days, 15 days and 100 days, respectively, after unilateral olfactory bulbectomy.

ipsilateral to the olfactory bulb lesion was markedly decreased in voltage immediately after the surgery in all animals, although EEG of the contralateral amygdala showed no change (Fig. 3). After a week or so, the reduced voltage of EEG in the ipsilateral amygdala was gradually restored, but never recovered its control level throughout the experimental period of approximately 1 month.

EEG changes after septal lesions

Effects on EEG of septal lesions were examined in 9 rats with permanent
electrodes implanted in the neocortex, amygdala and hippocampus. Immediately after the septum was bilaterally lesioned by electrocoagulation, the rats exerted hyperemotional responses to given stimuli. In this case, hyperreactivity such as jumping, flight and squeak responses was more prominent and attack response was less marked. This hyperemotionality gradually disappeared within 10 to 14 days after septal lesions. Accompanied with the changes in behavior, EEG of the neocortex changed to an arousal pattern consisted of low voltage fast waves, while the hippocampal theta activity was markedly decreased in voltage and desynchronized immediately after the lesions in all animals. In 3 out of 9 animals, spike activities of high voltage were also observed sporadically in the hippocampal

Fig. 4. Effect of bilateral septal lesions on EEG activity in the rat.  
A: control EEG. B, C and D: 1 day, 6 days and 10 days, respectively, after bilateral septal lesions.

Fig. 5. Effect of bilateral septal lesions on EEG activity in the rat.  
A: control EEG. B, C and D: 2 days, 4 days and 13 days, respectively, after bilateral septal lesions.
EEG (Fig. 4). These changes in the hippocampus continued throughout the experimental period of 1 month and never recovered the control pattern even after the septal hyperemotionality disappeared (Fig. 5). No change was observed in EEG of the amygdala after septal lesions.

**DISCUSSION**

Following bilateral olfactory bulbectomy, the rat exerted characteristic hyperemotionality which was somewhat different from that induced by septal lesions. Septal hyperemotionality appeared immediately after brain lesioning and disappeared within a period of approximately 2 weeks, while the hyperemotionality of the rat with bilateral olfactory bulbectomy (O.B. rat) appeared more gradually, reaching its maximum at 1 to 2 weeks after the surgery, and remained unchanged for at least 1 month of the observation period. In the O.B. rat, offensive aggression such as attacking a rod and muricide was more prominent but hyperreactivity was less marked. On the contrary, hyperreactivity was more prominent and offensive aggressiveness was less marked in the septal rat. These behavioral changes in the O.B. rat observed in the present investigation agreed well with the results reported by a number of investigators (Watson 1907; Vergnes and Karli 1963; Myer 1964; Ueki et al. 1972a).

Accompanied with the appearance of hyperemotionality, low voltage fast waves increased in spontaneous cortical EEG and the hippocampal theta activity was markedly synchronized after bilateral olfactory bulbectomy. This EEG arousal pattern in the O.B. rat was much more marked than that seen at an awake state in intact rats, in good parallel with behavioral arousal. The same arousa pattern was also observed in cortical EEG of the septal rats. On the contrary, the hippocampal theta activity was completely desynchronized and hippocampal EEG turned to low voltage fast activity immediately after septal lesions. This might be rational, since it was suggested that the development of synchronized theta waves in the hippocampus was dependent on the activity of the septum (Petsche et al. 1962; Stumpf et al. 1962).

The most remarkable change was observed in EEG of the amygdala after olfactory bulbectomy; i.e. the voltage of EEG in the ipsilateral amygdala was markedly reduced immediately after bulbectomy. Such a change was never induced by septal lesions.

It has been known that the afferent fibers originating from the olfactory bulb project directly in the amygdaloid complex as well as in the prepyriform cortex through the lateral olfactory tract (Adey 1959; Powell et al. 1965). It is therefore reasonable that the olfactory bulbectomy immediately alters the activity of the amygdala. Callen and Colle (1965) and Callen et al. (1969) have suggested that the olfactory bulb inhibits the activity of the prepyriform cortex, because the cellular activity of the prepyriform cortex is increased by olfactory bulbectomy. It is also conceivable that similar mechanisms may exist on the activity of the amygdala, and olfactory bulbectomy causes an increase in amygdaloid activity.
EEG changes in the amygdala observed after olfactory bulbectomy in the present study might reflect such changes in activity of the amygdala.

It has been well known that the limbic system plays an important role in regulating emotional behavior of animals (Klüver and Bucy 1937; Papez 1937), and King and Myer (1958) have suggested that, in general, the septum functions as an accelerator for the emotional center in the hypothalamus. It seems thus likely that olfactory bulbectomy results in activation of the amygdala and in turn induces hyperemotionality in rats.

However, the hyperemotionality of the O.B. rat appears rather slowly in spite of the fact that the amygdaloid EEG is altered immediately after bilateral olfactory bulbectomy. It is therefore difficult to explain this hyperemotionality merely through the activity changes in the amygdala. Changes in function of the other brain structures connected with the olfactory bulb should also be taken into consideration in order to explain the neural mechanism of this hyperemotionality. Nurimoto et al. (1974) have suggested that dysfunction of the olfactory tubercle may be more important in the development of hyperemotionality after olfactory bulbectomy. The olfactory bulb also sends afferent fibers to the anterior olfactory nuclei which are connected with the olfactory tubercle, septum, hypothalamus and further with the midbrain structures through the medial forebrain bundle (Natua 1958). In the present study, some parts of the anterior olfactory nuclei were also lesioned with olfactory bulbectomy. It might be, therefore, quite possible that the activity of these structures was somehow changed by this lesion. It is suggested that the marked arousal pattern of the neocortical and hippocampal EEG induced by bilateral olfactory bulbectomy results from activity changes in the hypothalamic (Gellhorn 1961) as well as in the mesencephalic reticular activating system (Moruzzi and Magoun 1949), but unilateral ablation of the olfactory bulb is not sufficient to cause such changes.

However, it should be also emphasized that the marked changes in amygdaloid EEG induced by olfactory bulbectomy may still indicate the importance of amygdaloid activity in the occurrence of hyperemotionality in the rat.

References


