AUTONOMIC RESPONSES ACCOMPANYING CONDITIONED
AND UNCONDITIONED ALIMENTARY REACTIONS IN AMYGDALO-
HYPOTHALAMICALLY LESIONED DOGS

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Abstract. Lesions in the amygdalo-hypothalamic system decreased conditioned and unconditioned alimentary responses in five dogs. During three postoperative months no improvement was observed. The introduction of the new food reward (chosen for each dog as the most preferred) produced a sudden or progressive increase of conditioned and unconditioned alimentary performance. In the cardiac responses accompanying postoperative conditioned and unconditioned alimentary reactions, the lack or diminution of the decelerative response during food consumption and a less pronounced postoperative acceleration of heart rate during conditioned stimuli were observed. Conditioned and unconditioned salivary responses decreased postoperatively and the pattern of the salivary responses varied individually. The results support the hypothesis that damage of amygdalo-hypothalamic system produces a decrease of the hedonic values of the usually used food rewards. Decrease or lack of the decelerative cardiac response during eating may reflect this ahedonic state.

INTRODUCTION

Recently, we have demonstrated that the beginning of eating is characterized by heart rate (HR) acceleration and inhibition of salivation. In the course of eating HR progressively decelerates and salivation in-
crease (18, 19). The patterns of these autonomic responses are presumed to reflect both the excitatory (drive inducting) and the rewarding components underlying the processes of reinforcement. It means that these patterns are closely connected with the motivational state of the organism. It was well established that an essential role in motivational behavior played the amygdalo-hypothalamic system, which was best demonstrated with respect to alimentary behavior. In dogs, dorsomedial amygdala lesions, similarly to lesioned of the lateral hypothalamus, produce a syndrome of aphagia or hypophagia and impairment of instrumental alimentary responses (6–10, 31). Also, conditioned and unconditioned salivary responses are changed in amygdalar (26) and in hypothalamically lesioned animals (32, 35). It is known from several studies that lesions or stimulation of the amygdala or hypothalamus produce changes in cardiac activity in various species, like rats, rabbits and dogs (4, 15, 33, 34, 41, 42). Therefore, it seems worthwhile to examine the effect of amygdalo-hypothalamic lesions causing a decrease of alimentary motivation on definite patterns of salivary and cardiac responses accompanying alimentary conditioned stimuli and alimentary consummatory acts.

METHODS

Subjects: Experiments were performed on 7 dogs in a sound-proof conditioned reflex chamber. All dogs had a chronic parotid gland fistula performed by Soltysik and Zbrożyna method (40). Heart rate (HR) was recorded through three electrodes fastened on a leather belt fastened around the body of each dog, near the heart.

Training. Instrumental conditioned responses were established to 1,000 Hz tone of 10s duration. The dogs were required to press a lever to the CS, which was reinforced by food immediately after instrumental performance and followed by CS offset. As the reinforcement, a constant portion of bread-powder mixed with boiled meat (about 50 g) was used. Each experimental session consisted of 8 trials. Intertrial intervals were 2–3 min and 5 sessions were conducted in each week.

Surgery. When dogs reached the criterion (90% of correctly responses in 10 consecutive sessions) the bilateral coagulations of the medial amygdala and/or lateral hypothalamus were performed using unipolar steel electrodes 0.5 mm in diameter, coated with enamel except for 0.5 mm at the tip. Lesions were produced by passing 3.5 mA anodal current for 2 min on points choosen according to Lim's et al. atlas (24).
Postsurgery training. The experimental sessions were resumed on the 10th day after operation, and conducted only 3 times per week, because operated animals did not accept the experimental situation (they either did not eat, or ate rarely in the reflex chamber, they barked and bit). Their performance of CRs was greatly deteriorated. However, the dogs were not retrained by the method of passive movements (17), on the contrary they received food after CS offset, in spite of the lack of CR, according to Wyrwicka method (46).

Food-preference tests. Three months after the operation food preferences were tested. The dogs were allowed to eat the standard food (bread powder mixed with boiled meat), fresh meat, cheese, smoked fish, cooked egg or fresh egg mixed with sugar. Each of these foods was presented in a separate small bowl. The latency of choice and the amount of the consumed food determined food preferences. This test was repeated twice: first, in the reflex chamber and later in another room.

When food preferences had been established, 10 experimental sessions were performed with the most preferred food (according to individual preferences) as reinforcement. In the case when it was not possible to established food preferences, or when the dog consequently failed to eat in the reflex chamber in spite of the wide range of food “cafeteria”, 10 experimental sessions with water as reinforcement were performed in water-deprived dog, fed with salty soup.

Measurements and data analysis. Both conditioned instrumental responses and unconditioned alimentary responses were recorded during: (i) 10 preoperative sessions, (ii) 10 postoperative sessions, (iii) the next 10 postoperative sessions, (iv) 10 sessions before change of food reward, and (v) 10 sessions with the most preferred food or water as reinforcement. In 5 dogs (from 7) in which lesions produced a deterioration of alimentary behavior, cardiac and salivary responses were measured in the following experimental periods: 2 preoperative periods of 10 sessions each, and 2 successive postoperative periods of 10 sessions each. Heart rate (HR) and salivary responses were both measured during each period: during 10 s of CS duration and during 10 s before CS onset. To determine the magnitude of HR and salivary responses during food consumption (US), the duration of each US was divided into three equal intervals and the heart beats and the drops of saliva occurring in each of them were counted. Finally, HR and salivary responses were measured during
postconsummatory licking which was observed until 10 s elapsed without any licking. HR was measured by counting the number of R-waves in the above described intervals. The mean HR for each dog was calculated from 10 trials and then converted to beats per minute. Salivary responses were measured by counting the number of drops of saliva in the same intervals during 10 trials and then converted to drops per 10 s. Additionally, in 3 dogs HR was measured during surgery before and 10 and 30 min after coagulation.

Analysis of variance (25) and Duncan tests (5) were used for statistical evaluation of the experimental data.

Histology. When the experiments were completed, the dogs were deeply anesthetized and perfused intracardially with isotonic salina followed by 10% formol-saline solution. The brains were removed and fixed in 10% formol-saline for several days. Transverse 50 μm sections were subsequently made with cryostat. The sections were stained by either Klüver or Nissl method.

RESULTS

Anatomy. We present only the results obtained in 5 dogs with extensive lesions of hypothalamus and/or amygdala. The criterion used to choose these dogs from the larger group of 7 dogs was the presence of obvious symptoms of alimentary disturbances and not the dimensions of lesions, which were different in particular dogs. The extent and localization of their amygdalo-hypothalamic damages are shown in Fig. 1.

Fig. 1. Schematic localization of amygdalo-hypothalamic lesion in individual dogs. Superimposed from histological slides within outline from about R-21 anterior (A) to posterior R-19 (P).
They involved mainly dorsomedial part of amygdaloid complex (n. medialis, area anterior) and lateral part of hypothalamus (n. lateralis, n. entopeduncularis) although in some cases other structures were more or less touched (i.e., internal capsule, optic tract, thalamus). In 2 dogs (P-5, P-8) amygdala was spared, but in 3 other dogs lesions involved both amygdala and hypothalamus.

**Alimentary behavior.** On the 10th day after operation 4 dogs were still hypophagic (P-1, P-4, P-5, P-8). In one dog (P-6) the symptoms of hyperphagia appeared, but its character was fluctuating (hypophagia mixed with hyperphagia), (Table I). Although 1 month after operation only two dogs were still hypophagic (P-4, P-5), all dogs during the first postoperative month demonstrated aversion to the experimental situation (escape behavior, barking, biting, aggression and motor excitement). Deterioration in alimentary behavior consisted in the decrease of URs, and prolongation of URs's latencies. Often the dogs failed to consume all portions of food. Also the licking after food consumption was shortened when compared with that before operation. Those symptoms varied among the dogs. In 3 dogs (P-1, P-4, P-5) these alterations were still observed 3 months after operation, when the dogs were tested for food preferences. Dog P-6 lost the negative attitude towards the experimental situation, but it was not especially interested in food consumption (it consumed successively all kinds of food, but did not show any food preferences). However, when tested for food preferences in a different chamber, four dogs chose and ate the preferred foods. The food preferences varied individually and we have not established any group tendencies. Only one dog (P-1) failed to eat also in a different chamber. It did not eat the salted soup either, thus we have not conducted the experiments with water as reinforcement in that case. In all four dogs the change of reinforcement produced an increase of URs and a sudden

<table>
<thead>
<tr>
<th>Dogs</th>
<th>During 10 days after operation</th>
<th>On 10th day after operation</th>
<th>1 month after operation (mean from 5 days)</th>
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<tbody>
<tr>
<td>P-1</td>
<td>63</td>
<td>63</td>
<td>104</td>
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<tr>
<td>P-4</td>
<td>18</td>
<td>44.5</td>
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<td>P-5</td>
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<td>P-6</td>
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<td>145</td>
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<td>P-8</td>
<td>80</td>
<td>75.5</td>
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*TABLE I*

Postoperative food intake (in percent of preoperative level)
appearance or increase of instrumental CRs. (Fig. 2). An analysis of variance performed for alimentary URs (Periods X Subjects) with raw scores transformed to $\sin \sqrt{P}$ (which corrects the distribution of percent-

gages to better estimate homogeneity of the scores and allow use of analysis of variance technique) revealed a significant effect of Periods ($F_{4/19} = 12.36, P < 0.001$). Also an analysis of variance for alimentary CRs (Periods X Subjects) revealed significant effects of Periods ($F_{4/19} = 26.69, P < 0.001$). Duncan tests indicated that in both types of alimentary behavior (URs and CRs) their postoperative decrease was significant ($P < 0.01$) and their increase after change of reinforcement was significant too ($P < 0.05$ for URs and $P < 0.01$ for CRs). In the opposite, during 3 consecutive periods after operation the performance of URs and CRs remained on the same level (differences not significant).

**Autonomic responses during food consumption.** In all dogs patterns of autonomic responses accompanying URs differed from those observed before operation. Cardiac responses (Fig. 3) after operation were characterized especially by the weakening of decelerative responses during food consumption (P-1, P-6, P-8), or even by the lack of decelerative responses (P-4). Other changes of cardiac responses during eating varied individually and were more dependent upon the preoperative pattern. For example in dog P-1 before operation HR accelerated greatly of the beginning of food consumption, but after operation it decelerated at the start of eating. On the other hand, two other dogs demonstrated totally different changes (P-4, P-6). In dog P-8 the preoperative initiation of
eating was accompanied or by HR acceleration or by HR deceleration, and after the operation, no changes at the start of eating were observed. Postoperative cardiac pattern during postconsummatory licking differed also from the preoperative one in most of the dogs, but the direction of these changes varied individually too.

The dynamic of salivary responses during food consumption was changed after operation in all dogs (Fig. 4), but the direction of these changes varied individually. The only change which appeared in most dogs was an increase of salivation during postconsummatory licking (P-1, P-4, P-6, P-8). Only in one dog (P-6) the general decrease of salivation during the first postoperative period was observed.

**Autonomic responses during exposition of CSs.** Accelerative cardiac responses to the CSs remained after operation in all dogs, but the magnitude of this acceleration decreased (Fig. 5). In three dogs (P-1, P-4, P-5) the general increase of cardiac activity was observed after operation, when checked in the reflex chamber. Postoperative salivary responses

Fig. 3. Cardiac responses recorded in individual dogs.

Fig. 4. Salivary responses in individual dogs.
Fig. 5. HR during CS exposition (crossed bars) compared with HR before CS (open bars) in individual dogs. A and B, two successive preoperative periods; C and D, two succeeding postoperative periods. Each bar represents mean value from 10 trials. Arrows indicate the operation.

to the CSs varied within the group (Fig. 6). In three dogs (P-5, P-6, P-8) conditioned salivation to CSs decreased after the operation. In dog P-4 salivary responses decreased only in the second postoperative period. Whereas in dog P-1 salivary responses increased in the second postoperative period.

Individual differences in the direction and magnitude of these changes were probably the cause of the fact that two analyses of variance (for salivary and cardiac responses), performed globally for all experimental periods and all session periods and subjects, have not revealed any statistically significant influence of the operation upon these responses.

Cardiac responses during surgery. Cardiac activity was checked immediately after the coagulation in three dogs and in two of them (P-4 and P-5) cardiac activity was unchanged. In the third one (P-6) coagulation produced a general deceleration of HR; (decrease from 175 to 152 beats per 1 min).
DISCUSSION

In dogs lesions of dorsomedial amygdala and/or lateral hypothalamus produced a clear deterioration of their alimentary behavior. The dogs did not eat (or ate rarely) in the experimental situation, they did not perform instrumental CRs for food and they demonstrated aversion to the experimental situation. These data agree with the previous findings (6–10, 31). The deterioration of alimentary behavior remained almost unchanged during 3 postoperative months. The change of food reinforcement to preferred food produced sudden or progressive improvement of the unconditioned and conditioned alimentary behavior. Our data support the earlier findings obtained on rats with lateral hypothalamic lesions, where it was demonstrated that a decrease of instrumental responses to food and unconditioned food consumption could be reversed by a change of diet to the more preferred food (1, 43). So far it was demonstrated only that the change of reinforcement facilitated the restitution of instrumental CRs in the neurotic dog (48). As the changes of food preferences in hypophagic hypothalamic and amygdalar dogs (7, 9, 31) were previously observed, the actual data are not surprising. It was more important that the restitution of the instrumental CRs could be so rapid when the preferred food was given. It means that amygdalar and hypothalamic lesions did not disrupt the connections between CS, CR and reward. The mnemonic processes seemed to be strongly preserved, because reinforcement by food of CS exposition, in spite of the lack of instrumental performance during 3 postoperative months, has not extinguished the CRs. The postoperative decrease of instrumental CRs in our dogs could be understood as a secondary symptom resulting from the impairment of unconditioned alimentary reflexes, due to the decrease of reinforcing value of food reward. Unfortunately we could not answer the question what was the cause of this diminished value of reward. The postoperative changes in food of preferences seems to be the major reason.

Contrary to the instrumental CRs, both cardiac and salivary responses seemed to be partially independent from the unconditioned food consumption. For example, in dog P-4 the salivary conditioned reflexes were not changed in the first postoperative period in spite of the animal's clear negativism to food. They dropped markedly only in the second postoperative period. Similar delayed effect of operation was observed by other authors (49). In the other dog P-8 the salivary conditioned reflexes decreased after operation (when compared with the period directly proceeding operation) and remained on the same level during both postoperative periods, in spite of the fact that only in the first postoperative period its unconditioned alimentary reflexes were greatly
deteriorated. In dog P-5 the salivary responses during CS exposition were preserved, although the animal did not eat during all postoperative periods. The same symptoms were observed earlier in the amygdalar dog (26). The decrease of salivation was not parallel to the decrease of food intake, because it appeared in dogs with pronounced aphagia and in dogs with only a small decrease of food intake. Similar independence of salivary reflexes from alimentary unconditioned reflexes was also observed in dogs after ablation of alimentary taste area i.e., the area comprising anterior composite gyrus and the most anterior part of coronal gyrus (47). As it is known that aversive alimentary stimuli produce the most abundant salivation (11, 29), it is probable that hypernormal conditioned salivation observed in some of our dogs could be a result of excitation of aversive mechanisms, since they dogs demonstrated aversion towards food. Therefore, it seems that conditioned salivary responses could not be a good index of hunger drive, which was already indicated by Soltysik (39). However, it should be stressed that different lesions may differently contribute to the changes in salivation in individual dogs.

Accelerative cardiac responses to the CSs decreased postoperatively in all dogs, but they were preserved, although it could be hardly explained by the activation of hunger drive. It seems probable that HR acceleration to the CSs was due to the excitation of aversive mechanisms. It may be due as well to a slow extinction of cardiac conditioned responses as well (12).

It is worthwhile to draw attention to autonomic responses accompanying unconditioned food intake. Their postoperative changes showed some interesting tendencies, especially in the case of cardiac responses. The dominant characteristic was the lack or weakening of the decelerative response during food consumption. As it was suggested previously (18–21), this decelerative cardiac response seemed to be connected with the hedonic states of the organisms, resulting from pleasant, sensory qualities of reinforcement. Thus, the weakening or absence of this response during eating may reflect the hedonic state of lesioned dogs. This interpretation could be supported also by changes in cardiac responses during the first period of eating. These changes indicated that the beginning of eating was often not associated with emotional excitement which normally occurs in this period (18, 19).

It is of interest to note that postoperatively, the decelerative cardiac responses accompanied by abundant salivation appeared often when food had been consumed. It could be due to the prolongation of latencies for these responses (as the result of an alteration of neuronal connections), or more probably to the changes of motivational processes
accompanying food consumption. It should be stressed that after operation the manner of food consumption was often "compulsive". If as a rule the dogs ignored food, sometimes they rushed at it and ate voraciously although they were not expecting food. Also, they did not seem to enjoy food consumption. These observations suggest that the motivation underlying these alimentary behaviors was different from the normal alimentary motivation. It could be due, for example, to a sudden eruption of hunger drive which dominated the consummatory, gustatory responses. But also another explanation could be possible. "Compulsive eating" was described in rats, in the compartment where previously they received painful electric shocks (45). This compulsive eating was explained as reducing the conditioned anxiety. Although we lack the data which could explain with certainty the motivational processes underlying "compulsive-like eating" in our dogs, it is important that the autonomic responses accompanying this type of eating differed from normal ones, before operation. Therefore, our data support the hypothesis that autonomic responses accompanying rewarding, consummatory acts seem to reflect their motivational contents (18-21).

It is important to mention that all postoperative changes described above (at least in cardiac responses) were not probably produced by a direct damage of the cardiovascular system, since no changes were observed when the cardiac activity was checked immediately after coagulation. That testifies rather of their emotional character and simultaneously supports the hypothesis that the damage of the amygdalo-hypothalamic system produces emotional changes which can be associated with the decrease of the hedonic value of reward (3, 6, 8, 10, 37).

Two questions still arise. First in which way the complicated processes of evaluation of the rewards' hedonic value take place in these subcortical centers of emotions. Second, which are the relationships between these processes and autonomic responses. It is known that changes in the cardiovascular system could be registered in these structures through their ascending neuronal connections from the lower centers of cardiovascular control (30, 44). On the other hand, the amygdala and hypothalamus could influence directly the cardiovascular system through their projections to the spinal cardiovascular centers (2, 28). Therefore a disruption of projections from cardiovascular centers to the amygdala and hypothalamus could eliminate the supply of informations concerning cardiovascular activity to these structures. According to James–Lange's theory (14), changes in the autonomic system could be a basis for emotions. Thus, it is probable that the lack of informations concerning autonomic responses, caused by the damage of the amygdalo-hypothalamic system, could result in a deterioration in
processing of the evaluation stimuli with regard to their hedonic and biological values. On the other hand, the activation of the additional subcortical mechanisms of cardiovascular control was no longer possible, which could also contribute to the processes of hedonic evaluations.

Determination of the autonomic and amygdalo-hypothalamic contributions to the processing of hedonic evaluations is an important problem and needs further studies, the more so as the decrease of alimentary behavior and changes of food preferences appearing after amygdalo-hypothalamic lesions seem to be of an emotional rather than basic gnostic character. The possibility of impairment of either gustatory or olfactory discrimination, postulated by other authors (16, 36), is doubtful, since after the lesions most dogs differentiated foods. Similar changes of food preferences without disturbance of sensory inputs were observed also in amygdalar cats (23, 47). Strong olfactory (22, 38), and gustatory (13, 27) inputs to amygdala and hypothalamus do not provide the satisfactory explanation in which way the discriminated foods after amygdalo-hypothalamic lesions became rewarding or aversive.

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