Effects of Amygdaloid Lesions upon Septal Hyperemotionality in the Rat

A striking increase in emotional behavior, manifested by violent attack or flight reactions in response to previously neutral stimuli, has been reported to result from experimental damage to the septal region of the forebrain in the laboratory rat (*1, 2*). A behavioral shift in the opposite direction occurs following lesions of the amygdaloid nuclei. Karli (*3*) and Woods (*4*) report that damage of this complex in the aggressive and unmanageable wild rat results in tameness and greatly lowered emotional reactivity.

The present investigation (*5*) was designed to obtain further information concerning the relative roles of the septal and amygdaloid regions of the brain in affective behavior by combining, in the same animal, lesions which produce opposite behavioral extremes. Thirty male hooded rats of the Lashley strain were used in this study, and the behavior of each animal was evaluated daily on a rating scale of emotionality. The scale was adapted, with certain modifications, from one used by Brady and Nauta (*1*) and contained the following six components: (i) attack or flight reaction to a rod presented visually before the rat's snout, (ii) startle reaction to a light tactual stimulation, (iii) resistance to capture, (iv) muscular tension and resistance to handling, (v) vocalization during testing, (vi) urination and defecation during testing. Each of these components was evaluated on a scale from 0 through 5, and an over-all daily score was assigned each animal by adding up the scores obtained for each component of behavior. All brain lesions were bilateral and were produced electrolytically with a unipolar electrode inserted by means of a stereotaxic instrument.

In experiment 1 the effects of amygdaloid damage imposed upon an already hyperemotional septal animal were evaluated. Ten animals designated septal-amygdaloid and five rats designated septal-cortical were rated on the emotionality scale for 3 days preoperatively; then both groups were subjected to septal damage and rated for 3 days following the operation. The next day all 15 animals were reoperated; ten were subjected to amygdaloid damage, and the remaining five received cingulate and neocortical lesions as a control for the effects of brain damage per se. From Fig. 1 it is clear that following septal damage all animals showed a striking increase in emotionality over preoperative values ($p < .01$). Further, the effect of an amygdaloid lesion imposed upon a septal preparation is to reduce suddenly the hyperemotionality observed during the 3 days following the septal operation ($p < .01$). In fact, such animals appear to return approximately to their preoperative emotional levels. Those septal animals which received cingulate and neocortical damage in lieu of amygdaloid lesions showed the gradual decline in emotional reactivity reported in previous studies in which only the septal area had been destroyed (*1, 2*).

Since amygdaloid lesions were found to produce a profound reduction in the emotionality of septal preparations, experiment 2 was performed to determine whether amygdaloid lesions placed prior to septal damage would effectively prevent the appearance of septal hyperemotionality. In this experiment ten animals designated amygdaloid-septal and five rats designated cortical-septal were rated on the emotionality scale for 3 days preoperatively. The former group was then subjected to amygdaloid damage, and the latter group received cortical lesions. Following three more days of ratings, all animals were reoperated and subjected to septal damage. Figure 2 shows that neither the initial amygdaloid nor cortical damage had an effect upon emotionality. When septal damage was then imposed upon the control group of five cortical-septal animals, an increase in emotionality similar in all observable respects to that seen in animals receiving septal damage alone was manifested. In the case of the experimental group of amygdaloid-septal animals, amygdaloid lesions were successful in preventing the appearance of a full-blown septal hyperemotionality. However, these animals did show a gradual, but only partial, development of hyperemotionality, differing from both their own preoperative values and the values reached by the septal-cortical group in experiment 1 at the .05 level of confidence.

The brain of every animal was studied histologically. In general, damage was restricted to the structures for which the lesion was intended, and no other structures of the brain consistently suffered injury. All animals designated "septal" were found to have suffered severe destruction of the precommissural and supracommissural portions of the septal region. Those animals designated "amygdaloid" all sustained damage to the amygdaloid complex; most heavily and consistently damaged were the lateral and basal nuclei. In no case was there total destruction of the amygdala. The animals referred to as "cortical" controls were found to have lesions of the anterior cingulate and adjacent neocortex.

It may be noted that, in contrast to previous studies (*3, 4*), amygdaloid lesions alone did not reduce affectivity. We suspect that the rats used in the present experiment were so low in emotionality preoperatively that differentiation by the rating scale in the direction of lowered emotionality was not possible.

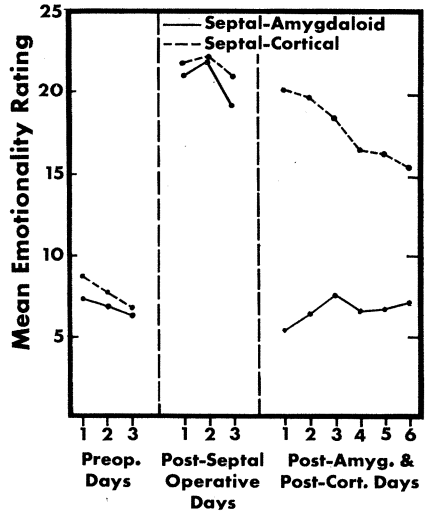

Fig. 1. Daily mean ratings of emotionality for septal-amygdaloid and septal-cortical operates.

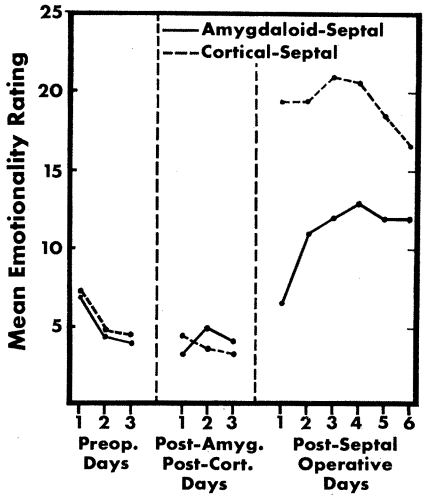

Fig. 2. Daily mean ratings of emotionality for amygdaloid-septal and cortical-septal operates.

Studies now in progress in this laboratory, in which rats with an initially high level of emotional responsivity are being used, indicate that this was probably the case, for such animals show a decided and sudden drop in reactivity following amygdaloid lesions. It is curious that amygdaloid lesions were able to produce total reduction of septal hyperemotionality in experiment 1, but only partial suppression in experiment 2; and although we are presently unable to offer a satisfactory explanation, it is conceivable that the effects of the amygdaloid lesions in experiment 2 have become partially dissipated by the time the septal lesion is placed, thus permitting a degree of hyperemotionality to appear.

The results of this study suggest that the septal region and amygdaloid nuclei may play reciprocal roles in the control of affective behavior. It appears possible that impulses from the septal and amyg-

daloid regions are directed toward some integrative level of the brain, probably the hypothalamus, since Bard and Mountcastle (*6*) have demonstrated that the hypothalamus is a critical center for the integration of emotional display. Furthermore, hypothalamic connections have been established for both the septal and amygdaloid areas. It appears that, in the rat, the septal area may normally act to "dampen" the hypothalamic activity associated with emotional states, while the amygdala may facilitate this diencephalic center.

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References and Notes

1. J. V. Brady, W. J. H. Nauta, *J. Comp. and Physiol. Psychol.* **46**, 399 (1953).
2. F. A. King, *J. Nervous Mental Disease* **126**, 57 (1958).
3. P. Karli, *Behavior* **10**, 81 (1956).
4. J. W. Woods, *Nature* **178**, 869 (1956).
5. Experiment 1 was carried out while the first author was on a U.S. Public Health Service predoctoral fellowship (MF-5490-C), and was submitted as part of his doctoral dissertation at Johns Hopkins University, under the direction of Professor C. T. Morgan. Experiment 2 was supported by a grant from the National Institute of Mental Health (M-1639) and carried out at Ohio State University.
6. P. Bard and V. B. Mountcastle, *Research Publs. Assoc. Research Nervous Mental Disease* **27**, 362 (1947).

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Radiation-Protective Effects of Yeast Extract and Yeast Ribonucleic Acid

Most of the work with biological substances in the field of radiation protection has been with proteins, amino acids, animal cells, and their extracts. Studies involving the administration of embryonic cells, viable spleen and bone marrow cells, either in the pre- or postirradiation period, have been voluminous and unequivocally show varying degrees of protection.

On the other hand, experience with plant substances and yeasts in particular has been quite limited. It has been demonstrated by Jaraslow *et al.* (*1*) that the administration of an autolysed yeast extract to rabbits is capable of protecting the postirradiation response to certain immunologic stimuli. Hollaender and Doudney (*2*) have demonstrated that irradiated *Escherichia coli* grown aerobically in nutrient broth recover from x-ray effects to a considerable degree if they are plated after irradiation on agar containing yeast extract. In studies designed to evaluate the role of properdin in radiation protection, Ross *et al.* (*3*) studied postirradiation survival of rats and mice after injections of zymosan. A moderate protective effect was demonstrated. Because of the suggestion from these studies that yeast and yeast extracts might have radiation-protective properties, an evaluation study in rodents was performed.

Autolysed yeast extract was prepared according to the method described by Jaraslow *et al.* (*1*) in which dried brewer's yeast was incubated with isotonic phosphate buffer at *p*H 7.4 at 37°C for 12 hours and then centrifuged at 20,000 *g* for 30 minutes. The clear supernatant autolysate was used for injection. For each experiment fresh yeast autolysate was prepared. Within from 15 to 30 minutes prior to irradiation, 200-g Sprague-Dawley rats were injected either intravenously or intraperitoneally with 1.0 ml of the autolysate.

In the first experiment, a total of 27 injected rats were compared with a group of 50 uninjected controls. In the second experiment, 15 eleven-week-old C_3H mice were injected either intraperitoneally or intravenously with 0.5 ml of the yeast autolysate. The survival rate of these mice was compared with that of 105 uninjected controls. Thirty minutes after injection the rats were placed in a polyethylene box and irradiated in pairs so that each animal received a total body dose of 900 r of x-ray over a period of 17 minutes. In a similar fashion the mice were placed in a plastic container and given 700 r of total-body radiation of x-ray over a period of 20 minutes. The number of surviving animals then was checked at daily intervals during the 30-day postirradiation period. All but one of the uninjected control rats were dead by the 13th postirradiation day, and this animal succumbed on the 21st day. On the other hand, 11 of the 27 injected rats (41 percent) were alive on the 30th day after irradiation. Four of the 15 yeast autolysate-injected mice (27 percent) were alive on the 30th day after irradiation, whereas all of the uninjected control mice were dead by the 14th postirradiation day. In both groups of animals the results obtained with intravenous and intraperitoneal injections were comparable. It also was noted that even the nonsurviving rats and mice lived longer than did the irradiated controls.

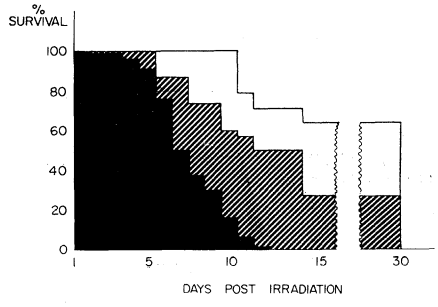

Fig. 1. Percent survival of three groups of C_3H mice following 700r total body irradiation. (a) Solid black area—105 controls (b) shaded area—15 yeast autolysate injected and (c) clear area—14 yeast RNA injected.

The results of these experiments indicated that the crude yeast autolysate provided a moderate degree of radiation protection to lethally irradiated animals. Yeast autolysate is extremely rich in ribonucleic acid (RNA), and there is good evidence that RNA is essential for protein synthesis. Studies with transplanted, irradiated nuclei of amoebae (*4*) suggest that irradiated cytoplasmic constituents of these cells are deleterious to normal nuclear function. Daniels (*5*) has suggested that non-nucleated cellular components of normal amoebae are capable of restoring nuclear function to irradiation-damaged cells. The high RNA content of cellular cytoplasm of many protective tissue extracts made it seem possible that the RNA of the yeast autolysate was its most important protective constituent and prompted investigation of the radiation-protective effects of this nucleic acid.

A group of 14 eleven-week-old, C_3H mice were injected intraperitoneally with either 10 or 20 mg of a commercial preparation of yeast RNA containing less than 1 percent protein (Schwarz Laboratories). The RNA concentration of the solution used for injection was 2.0 g/100 ml. Fifteen to 30 minutes later these animals were exposed to 700 r of total-body irradiation. Nine of the 14 injected mice (64 percent) were alive at the end of 30 days. These results are to be compared with no survival in the control group of 105 mice, and with 27 percent survival in the group injected with crude yeast autolysate (Fig. 1). There is some indication from these data that the dose of RNA may be of importance since only one of the five mice that died received the 20-mg dose.

In each of the three cases, a chi-square test was applied to determine whether the difference in survival rates between the two groups is significant. The resulting *P*-values are less than 0.001 in all three. These results indicate that the preirradiation injection of a crude yeast autolysate exerts a moderate radiation-protective effect in rats and mice. Postirradiation survival is considerably enhanced with a commercial yeast-RNA preparation. It is of interest that a plant extract devoid of viable cells and rich in RNA is capable of exerting protective effects to a degree comparable to that of many mammalian cells and tissue extracts. Preliminary results indicate that RNA, or a substance associated with it in the yeast autolysate, may be the principal radiation protective factor (*6*).