Table 1. Noradrenaline released by incubation of granules from bovine splenic nerves with reserpine (lyophilized reserpine phosphate) or Serpasil solution. Suspension medium, 0.075*M* K-phosphate, *p*H 6.5 to 7.0. Incubation time, 30 minutes at 20°C.

Concentration (m*M*)	Noradrenaline left in granules (% of control)
Control solution	
	100
Reserpine solution	
0.025	95, 100
.075	76, 96
.125	66, 59
.175	52
.25	46, 33, 44, 37
.5	17
.625	26

It has been shown that reserpine causes a depletion of the catecholamines from the heart (*3*), from the hypothalamus (*4*), and from the suprarenal medulla (*5*). In the cat and the rat, denervation does not prevent the action on the adrenal medulla (*6*). With large doses of reserpine (5 mg/kg), noradrenaline was found to be released from the rabbit's heart even after section of the cord at C_1 and C_2 (*7*). Moreover, reserpine is active on the heart-lung preparation (*8*), and depletion of rat heart and intestine is not prevented by ganglionic blocking agents (*9*). These results suggest a direct action on the stores. The present report deals with the effect of reserpine on the release of noradrenaline from isolated granules from bovine splenic nerves.

Bovine splenic nerves were stripped of their sheath and a press juice was obtained by squeezing the nerves between nylon cylinders in the cold. The residue was washed with ice-cold 0.075*M* potassium phosphate of *p*H 7.5, 5 to 10 ml per gram of nerve. The combined press juice and washing fluid (*p*H 6.5 to 7.0) was centrifuged in the cold for 5 minutes at 1000*g*, and the sediment was discarded. The supernatant was centrifuged at 50,000*g* for 30 minutes at 3°C, and the sediment was resuspended in 0.075*M* potassium phosphate, *p*H 6.5 to 7.0. Reserpine was added to the phosphate buffer in various concentrations, and the suspension was gently agitated during incubation for 30 minutes at room temperature (20°C).

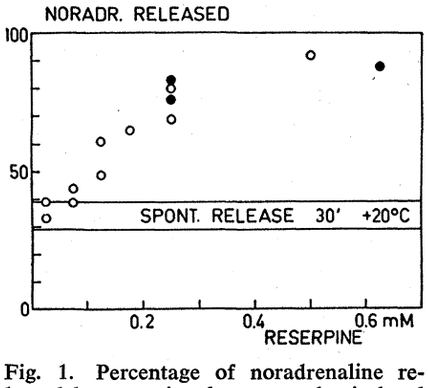

Fig. 1. Percentage of noradrenaline released by reserpine from granules isolated from bovine splenic nerves, sedimented, resuspended in 0.075*M* potassium phosphate, and incubated for 30 minutes at *p*H 6.5 to 7.0 at 20°C. Open circles, lyophilized reserpine phosphate; solid circles, Serpasil in ampule solution.

The reserpine preparations used were Serpasil (Ciba) and soluble lyophilized reserpine phosphate in the extract (*10*).

The controls were incubated with the solvents in the same way. Solutions made of the lyophilized reserpine phosphate in 0.075*M* potassium phosphate were stable at *p*H 6.5 to 7.0 at room temperature but showed precipitation at lower temperatures.

After incubation, the suspensions were recentrifuged and the amounts of noradrenaline in the supernatant and in the sediment were estimated by a fluorimetric technique (*11*). The noradrenaline in the sediment was quantitatively released by the addition of 1 ml of 1-percent metaphosphoric acid, and the amount in the supernatant was estimated after sedimentation of the precipitated granules.

The effects of various concentrations of reserpine on the release of noradrenaline from the granules are shown in Table 1. From the table it can be seen that approximately one-half of the noradrenaline present in the control samples remains after incubation with 0.175 m*M* reserpine and that less than 30 percent remains after incubation with 0.5 to 0.625 m*M* reserpine for 30 minutes at room temperature.

Figure 1 shows the release of noradrenaline as a percentage of the total amount present in the primary sediment. As seen in Fig. 1, about 90 percent of the total noradrenaline is released in the presence of 0.5 m*M* reserpine during the experimental conditions, as against 29- to 39-percent release in the controls during the same period. No releasing action was found on incubation of the granules with Serpasil solvent.

The experimental results reported here (*12*) are in agreement with the assumption that reserpine depletes the transmitter in adrenergic nerves by acting directly on its stores. The results also give evidence that the effect may appear even when reserpine is allowed to act on isolated transmitter granules during a brief period, provided the concentration is sufficiently high. Lower concentrations acting over longer time periods have not been tested, in view of the considerable spontaneous release at room temperature. At lower temperatures, on the other hand, reserpine was found to have little or no action, partly as a result of diminished solubility.

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Interhemispheric Effects of Cortical Lesions on Brain Biochemistry

Abstract. Unilateral lesions in the visual and somesthetic cortex of rat brain cause a slight but significant increase in the cortical cholinesterase activity in the contralateral hemisphere. There is some indication of strain differences in this effect. No change in cholinesterase activity is found in the subcortical brain. These findings may be helpful in understanding mechanisms of interdependence among brain areas.

A recurrent observation in the study of brain function is the apparent unity of the brain despite the equally apparent differentiation—Flourens' *action commune* and *action propre*. Limited cortical injury can show widespread effects in behavior presumably controlled by cortical tissue far removed from the site of injury; and a specific behavior pattern which deteriorates after ablation of its cortical center will show recovery with the passage of time or with special training. The mechanisms behind these phenomena have never been made clear.

One possible avenue for these generalized effects may be biochemical. Since

we had found reliable relations between cortical cholinesterase activity and adaptive behavior (*1*), and since cholinesterase activity is a general characteristic of the cortex (high interareal correlations being the rule), we investigated whether a limited cortical lesion might alter cholinesterase activity throughout the brain. We have been unable to discover any reports of generalized effects of limited cortical injury on brain biochemistry in either experimental animals or in man. This report concerns an exploratory study in this area—an area which we believe should be of concern not only for brain physiology but also for the medical management of brain injury.

In the main experiment 27 littermate pairs of 110-day-old rats of the S_1 strain were used. In one animal of each pair a variable amount of cortical tissue in the visual and somesthetic areas of one hemisphere was cauterized under ether anesthesia. In 12 animals the left hemisphere was operated on; in 15 animals, the right. Approximately 25 days after the operation the experimental animals and their littermate controls were decapitated and their brains prepared for chemical analysis. The cholinesterase activity of the visual, somesthetic, and motor cortical areas of the intact hemisphere were determined separately for each experimental animal. For each control animal these three areas were analyzed in the hemisphere corresponding to that of its experimental littermate. In addition, the cholinesterase activity of the subcortex (defined as total brain minus the dorsal cortex) was determined for every animal. The analytical procedure, with an automatic titrator, has been reported previously (*1*). Cholinesterase activity is measured in terms of moles of acetylcholine ($\times 10^{10}$) hydrolyzed per minute per milligram of tissue.

To obtain some indication of the generality of the effect, 20 additional littermate pairs of rats of approximately the same age as the others but of three other strains were also tested. Of these, ten were from the S_3 strain and ten from the RDH and RDL strains. The S_3 strain was derived from the same parental stock as the S_1 strain. These two strains were developed by Tryon (*2*) through selective breeding for maze brightness (S_1) and maze dullness (S_3). The RDH and RDL strains came from a different parental stock and had been developed by Roderick (*3*) through selective breeding for high (RDH) and low (RDL) cortical cholinesterase activity.

The results are presented in Table 1. An analysis of variance of the S_1 data, taking into account the littermate design, produced two main findings. (i) The over-all cholinesterase activity of the visual, somesthetic, and motor areas in the hemisphere contralateral to the lesion was significantly higher for lesion than for control animals ($F = 9.25$, degrees of freedom = 1 and 26, $P < .01$). (ii) This effect did not differ significantly from one cortical area to another ($F = .64$, degrees of freedom = 2 and 52), and is thus generalized. Over-all, cortical cholinesterase activity in the lesion animals exceeded that in the control animals by 2.5 percent. This 2.5-percent increase can be of functional importance in view of the exceedingly small individual variability of cerebral cholinesterase activity (*4*). Actually, the increase represents about one-half of a standard deviation.

Table 1. Effects of cortical lesions on cholinesterase activity in contralateral cortex and in subcortex in various strains of rats. Missing *P* values indicate nonsignificant differences.

Item	S_1 (N = 27 pairs)		S_3 (N = 10 pairs)		RDH and RDL (N = 10 pairs)	
	Cortex	Subcortex	Cortex	Subcortex	Cortex	Subcortex
Lesion	68.9	167.2	62.2	148.0	60.1	140.2
Control	67.2	165.4	59.3	148.2	59.8	139.2
Difference	1.7	1.8	2.9	−0.2	0.3	1.0
P	<.01		<.05			

In the subcortex, in distinction from the cortex, the cholinesterase activity of lesion animals exceeded that of controls by 1.1 percent, and this difference was not significant.

Among the ten S_3 pairs, the results were similar to those of the S_1 animals; cortical cholinesterase was 4.9 percent greater in lesion than in control animals (significant at the .05 level of confidence). There was no difference in the subcortex. Among the ten pairs of RDH and RDL animals, cortical cholinesterase activity of the lesion animals exceeded that of the controls by only 0.5 percent, and the subcortical difference was only 0.7 percent; neither of these differences was significant. These observations, suggesting that strains may respond differently to cortical lesions, indicate the necessity of sampling several strains in further work.

The change in cholinesterase activity seems to be restricted to neural tissue similar to that in which the lesion is made. This is shown in our failure to find significant change in subcortical cholinesterase activity following cortical lesions and in the failure of Sperti and Sperti (*5*) to find changes in cerebellar cholinesterase activity following lesions in the cerebral cortex.

The observed increase in cholinesterase activity in the hemisphere contralateral to the lesion might be accounted for by either of the following hypotheses. (i) The increase is a direct result of the lesion. Such increase in cholinesterase activity would have functional consequences in tissue far removed from the lesion. (ii) The lesion results in a re-routing of brain activity, and the increase of function in the intact tissue leads to a compensatory increase in the chemicals important in neural transmission.

Our second hypothesis is congruent with recent reports from other laboratories. Research in Belgium (*6*) has shown that unilateral cortical lesions in cats lead to exaggerated electrical activity in the homologous area of the contralateral hemisphere. This is most clearly seen in the secondary responses to afferent stimulation. English work (*7*) indicates increases in cholinesterase activity in rat brain after increases in neural activity, while, conversely, American and Italian research (*8, 5*) indicates decreases in cholinesterase activity following decreases in neural activity.

We have further experiments under way which may make it possible to decide between these hypotheses. Whatever the nature of the effect, further investigation of it may provide increased insight into the mechanisms of interdependence among brain areas (*9*).

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