serum has been investigated in uninfected controls and animals infected with *Diplococcus pneumoniae*. No significant change of enzymatic activity was found in kidney, lung, sternum, femur, and spleen. As the infection progressed a decrease in activity was observed in the liver enzyme, while a marked increase occurred in small intestine. The enzymatic activity of the serum also increased during infection, but less strikingly. In organ-specific inhibition experiments it was shown that serum and intestinal enzyme behaved identically. The data support the concept that a major portion of serum AP may originate in the small intestine. Neither the infection-related increase in intestinal AP nor the fall in hepatic AP was dependent upon an intact adrenal gland in the presence of circulating glucocorticoid hormones.

1. Bodansky, O., Advance Cancer Res., 1961, v6, 1.
2. Gutman, A. B., Am. J. Med., 1959, v27, 875.
3. Hoffman, G. C., Lucich, V. M., Cleveland Clin. Quart., 1960, v27, 146.
4. Kenny, J. J., Moloney, W. C., Blood, 1957, v12, 295.
5. Kaplow, L. S., ibid., 1955, v10, 103.
6. Wachstein, M., J. Lab. Clin. Med., 1946, v31, 1.
7. Beisel, W. R., Blood, 1966, v18, in press.
8. Robinson, E., Hochman, A., Oncologia, 1964, v17, 55.
9. Mahy, B. W. J., Rowson, K. E. K., Salaman, M. H., Parr, C. W., Virology, 1964, v23, 528.
10. Garg, S. P., Sharma, G. L., Cornell Vet., 1965, v55, 190.
11. Slein, M. W., Logan, G. F., Jr., Bact., 1962, v83, 359.
12. Counts, G. W., Smith, I. M., Routh, J. I., Hazard, E. C., McTavish, J. F., Nature, 1961, v191, 783.
13. Shinowara, G. Y., Jones, L. M., Reinhart, H. G., J. Biol. Chem., 1942, v14, 921.
14. Fiske, C. H., Subbarow, Y., ibid., 1925, v66, 375.
15. Wolfson, W. Q., Cohn, C., Calvary, E., Ichiba, F., Am. J. Clin. Path., 1948, v18, 723.
16. King, E. J., Campbell, D. M., Clinica Chim. Acta, 1961, v6, 301.
17. Fishman, W. H., Green, S., Inglis, N. I., Biochim. Biophys. Acta, 1962, v62, 363.
18. ———, ibid., 1962, v62, 429.
19. Shambaugh, G. E., MacNair, D. S., Beisel, W. R., Am. J. Digest. Dis., 1967, v12, in press.
20. Lust, G. Fed. Proc., 1966, v25, 1688.

Received November 9, 1966. P.S.E.B.M., 1967, v124.

The Effect of Hypovolemia on Drinking in Rats With Lateral Hypothalamic Damage.* (31860)

EDWARD M. STRICKER AND GEORGE WOLF (Introduced by J. R. Brobeck)
Institute of Neurological Sciences, University of Pennsylvania, Philadelphia and The Rockefeller University, New York, N. Y.

Bilateral lesions of the lateral hypothalamic area of rats result in adipsia and aphagia. However, with appropriate post-operative care a partial recovery from these deficits may occur so that a rat with these lesions eventually begins to ingest sufficient water and food to maintain life(1,2). Such rats have been termed "recovered laterals." The physiological stimuli which induce drinking in recovered laterals are not known. Intracellular dehydration, the best known stimulus for normal thirst, is ineffective, as are simple water deprivation and hyperthermia(3). It appears that recovered laterals may drink water only while they eat dry food(3).

Rats normally show a close relationship between food and water intakes. Gregersen (4) has emphasized the pronounced reduction in blood volume (hypovolemia) during feeding which is caused by the rapid secretion of digestive juices. Since a decreased plasma volume can be an important stimulus

* This work was supported in part by Grants MH 00647 and AM 08196 from Nat. Inst. of Mental Health, and done during tenure of the first author as Postdoctoral Research Fellow of Nat. Inst. of Mental Health and of the second author as an Advanced Research Fellow of the American Heart Assn.

for thirst(5), it is conceivable that this is the basis for the drinking after recovery from lateral hypothalamic damage. The present study evaluates this possibility.

Method. Twenty-four male Holtzman Sprague-Dawley albino rats, approximately 90 days old and weighing 350 g at the beginning of experiment, were used. Bilateral lesions of the lateral hypothalamic area were produced in 12 rats by passing 1 ma anodal DC for 10 seconds through stereotaxically guided stainless steel electrodes which were insulated except for 0.5 mm at the tip. The remaining 12 rats served as controls, 6 without operation and 6 with sham operations that were done by bilaterally inserting the electrodes to just above the lateral area without passage of current (4 rats) or by unilaterally inserting an electrode into the lateral area with passage of current (2 rats). All control animals responded similarly to the experimental treatments and will be described as a single group.

Rats with bilateral lesions were tube fed and maintained on palatable liquid foods (*e.g.*, Metrecal, wet mash) for 2-6 weeks until they began to ingest Purina chow and distilled water *ad lib*. Four died during the recovery period, but all others survived and were able to maintain body weight on a Purina chow and distilled water regimen (Stage IV of recovery; (3)).

The experimental treatments were begun about one week after Stage IV of recovery had been reached. At 9 a.m. of the first test day, all rats were injected subcutaneously in the middle of the back with 5.0 ml of 1.0 M NaCl to induce hyperosmolarity, or with 5.0 ml of 10% polyethylene glycol (Carbowax, Compound 20-M; Union Carbide & Chemicals Co.) dissolved in 0.15 M NaCl to induce hypovolemia(5), or were given a sham treatment in which the needle was merely inserted and withdrawn. They were then deprived of food and water for 7 hours, after which a drinking tube containing distilled water was presented for one hour and the intake recorded. Each rat received each treatment only once, in a partially counterbalanced order, with 2 days intervening between successive treatments during which time Purina chow and water were available *ad lib*.

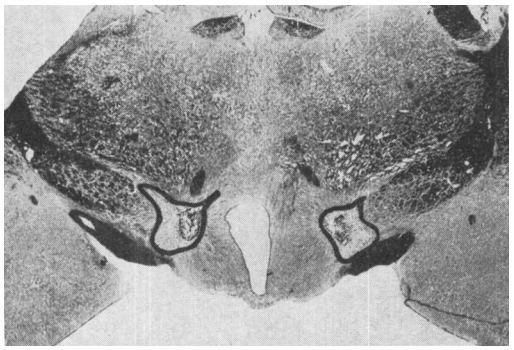

Fig. 1. Lesions representative of the size and placement of the lesions in the present study. They have been outlined in black ink.

To provide some information on the effects of bilateral lesions of the lateral hypothalamus on the renal aspects of the osmo- and volume regulatory processes, 6 recovered laterals and 6 control rats were kept in metabolism cages during the 7 hour food and water deprivation period following the above treatments. Urine was collected and its volume measured in a graduated tube attached to the base of each cage.

At the end of the study the recovered laterals were killed under anesthesia. They were perfused with 10% formalin and the placement of the lesions was determined by microscopic examination of stained brain sections.

Results. The lesions generally destroyed most of the tissue between the fornix and the internal capsule at the level of the anterior half of the ventromedial nucleus (Fig. 1). The lesions were about 1.0 mm in diameter dorsoventrally and mediolaterally and about 1.3 mm in diameter rostrocaudally. In most cases they were somewhat asymmetrically placed in the lateral dimension so that one of them encroached upon the fornix or the medial hypothalamic zone.

The physiological effects of the experimental treatments used have been discussed at length elsewhere(5,6). Briefly, subcutaneous injection of hypertonic saline solution leads to a hyperosmolarity of all body fluids with an intracellular dehydration, whereas injection of polyethylene glycol (PG) solution

TABLE I. Water Intake (ml) in Control Rats and in Rats with Lateral Hypothalamic Damage After Treatment with 1 M NaCl and 10% PG.

Recovered lateral	Treatment			Control rat	Treatment		
	sham	1 M NaCl	10% PG		sham	1 M NaCl	10% PG
1	.0	2.5	4.5	1	2.2	10.6	6.0
2	.0	3.0	.5	2	1.8	13.3	7.7
3	.0	1.0	.0	3	1.2	9.5	5.5
4	1.0	3.5	1.0	4	.5	10.0	6.2
5	.0	2.5	.0	5	2.0	8.2	6.2
6	.0	.5	5.0	6	.3	5.9	6.5
7	.0	.0	.0	7	.0	3.5	4.0
8	.0	.0	.0	8	1.0	5.5	4.0
				9	.5	7.0	5.5
				10	2.0	9.0	5.0
				11	.0	7.5	4.0
				12	1.0	10.5	6.5
Mean	.1	1.6	1.4	Mean	1.0	8.4	5.6

TABLE II. Urine Volume (ml) in Normal Rats and in Rats with Lateral Hypothalamic Damage After Treatment with 1 M NaCl and 10% PG.

Recovered lateral	Treatment			Control rat	Treatment		
	sham	1 M NaCl	10% PG		sham	1 M NaCl	10% PG
1	1.0	7.0	1.3	1	3.2	13.5	2.8
2	1.0	6.5	1.5	2	3.8	13.3	1.4
3	1.5	8.0	1.0	3	3.8	11.6	1.2
4	1.2	7.5	2.0	4	3.2	13.0	1.8
5	1.4	9.0	2.0	5	4.0	11.8	1.1
6	.9	10.5	1.0	6	3.8	12.6	2.0
Mean	1.2	8.1	1.5	Mean	3.6	12.6	1.7

causes a subcutaneous sequestration of an isosmotic plasma filtrate and consequently hypovolemia. Both these treatments elicit thirst in normal animals(6), as is illustrated by the data for the control rats presented in Table I (both p's $<.001$ by 2-tailed t test in comparison with sham treatment conditions).

In contrast, recovered laterals drank only a small fraction of the amount consumed by control rats after 1 M NaCl treatment ($p<.001$), after 10% PG treatment ($p<.001$), or after sham treatment ($p<.01$). It should be noted from the data presented in Table I that thirst was not completely absent in all 8 recovered laterals, since 6 drank some water in response to hyperosmolarity and 4 also drank in response to hypovolemia. On the other hand, only one rat (Rat No. 4) drank in response to the 7 hour deprivation period in the sham treatment condition and 2 (Rats No. 7 and No. 8) did not drink in response to any treatment. The lesions in the latter 2 rats were more symmetrically placed than were those in the other animals. However, the lesions in the 2 recovered laterals (Rats No. 1 and No. 6) that showed the least attenuation of drinking to PG treatment were indistinguishable from others of the series. In general, all lesions were somewhat smaller and less symmetrical than those described by Epstein and Teitelbaum(3) in rats which did not drink at all following treatment with hypertonic saline.

The effects of the various treatments on urine volume in recovered laterals and in control rats are illustrated in Table II. Like unlesioned rats, recovered laterals excreted a large volume of urine following 1 M NaCl treatment ($p<.001$, in comparison with sham treatment) and a small volume of urine following PG treatment. These effects were not as pronounced as in control rats ($p<.001$ for 1 M NaCl treatment). In addition, recovered laterals excreted a considerably lower volume of urine following sham treatment than did control rats ($p<.001$).

Discussion. In the present study, 4 recov-

ered laterals did not respond at all to the thirst-inducing effect of hypovolemia, and 4 others showed some impairment in drinking. These data argue against the hypothesis that decreased intravascular fluid volume provides the stimulus for spontaneous drinking in such rats. Since it is now clear that recovered laterals do not drink a normal amount in response to any of the usual stimuli which elicit thirst as part of a general homeostatic process (*i.e.*, hyperosmolarity, hypovolemia, and hyperthermia), it seems possible that certain peripheral cues are responsible for mediating drinking in these animals. A number of authors(7,8) have noted that the prandial drinking of recovered laterals is similar to that observed in salivarectomized rats. Since recent work indicates that recovered laterals have an impaired saliva production (9), it is possible that impaired salivary flow in combination with other oral stimuli arising from the ingestion of dry food are responsible for drinking in the recovered lateral. This hypothesis, first suggested by Epstein and Teitelbaum(3), seems the most likely explanation of the factors responsible for the drinking after recovery from lateral hypothalamic damage. The gradual onset of prandial drinking, also seen in salivarectomized rats, might then be attributed to the rats learning that water can effectively be used as exogenous saliva in facilitating the swallowing of dry food. However, it should be recognized that recovered laterals are not functionally equivalent with non-lesioned desalivate rats, since salivarectomized rats have been shown to respond normally to hyperosmolarity(8).

It is not clear whether aspects of the osmo- and volume regulatory processes other than drinking are impaired by lateral hypothalamic lesions. Andersson and McCann(10) have noted that low volumes of maximally concentrated urine are excreted by adipsic dogs (apparently in Stage III of recovery) following lateral hypothalamic lesions. This suggests that these lesions may not affect the regulation of ADH secretion. It is thus possible that the recovered lateral can maintain normal bodily hydration by enhanced renal water conservation in spite of a permanent hypodipsia. Of course, renal concentration mechanisms alone could not insure adequate hydration if water intake were sufficiently low, and the water intake of recovered laterals under *ad lib* feeding conditions is in fact often considerably lower than normals. In the present experiment, for example, the mean daily water intake of recovered laterals during the days intervening between treatments was only 17.9 ml, whereas the comparable intake of control rats was 28.0 ml. This low intake, and their unusually low volumes of urine excreted following sham or NaCl treatments, suggest that the recovered laterals may have been in negative water balance. In this regard, high serum sodium concentrations have been observed in rats that were hypodipsic following hypothalamic lesions (11). On the other hand, it remains possible that the recovered laterals in the present experiment were not dehydrated and that the reduced urine volumes actually reflect an adequate compensation for the hypodipsia. A more thorough investigation of water balance, body fluid composition, and renal function is required to determine whether renal mechanisms can maintain body fluid balance after lateral hypothalamic lesions.

Summary. Lesions in the lateral hypothalamus of rats led to impaired drinking responses to acute hyperosmolarity produced by 1 M NaCl injected subcutaneously and to acute hypovolemia produced by subcutaneous injection of 10% polyethylene glycol solution. Since recovered laterals drink while eating dry food, it was suggested that the lesions may disrupt structures mediating thirst arising from such stimula as hyperosmolariy and hypovolemia, but leave intact structures mediating drinking in response to peripheral stimuli such as dry mouth. Other evidence was presented which suggests that the renal aspects of osmo- and volume regulation are not impaired by the lesions.

1. Anand, B. K., Brobeck, J. R., Yale J. Biol. and Med., 1951, v24, 123.

2. Teitelbaum, P., Stellar, E., Science, 1954, v120, 894.

3. Epstein, A. N., Teitelbaum, P., in Thirst in the Regulation of Body Water, M. J. Wayner, ed., Pergamon Press, Oxford, 1964, 395-406.

4. Gregersen, M. I., Am. J. Physiol., 1932, v102, 344.
5. Stricker, E. M., ibid., 1966, v211, 232.
6. Stricker, E. M., Wolf, G., Physiol. Behav., 1967, v2, 33.
7. Epstein, A. N., Spector, D., Samman, A., Goldblum, C., Nature, 1964, v201, 1342.
8. Vance, W. B., Psychol. Monogr., 1965, v79, Whole No. 598.
9. Hainsworth, F. R., Epstein, A. N., Science, 1966, v153, 1255.
10. Andersson, B., McCann, S. M., Acta Physiol. Scand., 1956, v35, 312.
11. Stevenson, J. A. F., Welt, L. G., Orloff, J., Am. J. Physiol., 1950, v161, 35.

Received November 9, 1966. P.S.E.B.M., 1967, v124.

Anesthesia LXXI. Pharmacologic and Physicochemical Comparison of Flurothyl and Its Isomer.* (31861)

JOHN C. KRANTZ, JR., FRIEDA G. RUDO, AND CLAIRE K. LOECHER
Huntingdon Research Center and Department of Pharmacology, School of Medicine, University of Maryland, Baltimore, Md.

In our studies with fluorinated ethers the anesthetic, fluroxene(1) ($CF_3 \cdot CH_2\text{-}O\text{-}CH{:}CH_2$) and the convulsive flurothyl ($CF_3 \cdot CH_2\text{-}O\text{-}CH_2 \cdot CF_3$)(2) were developed and later used clinically. Recently we obtained the isomer of flurothyl, hexafluoroisopropyl methyl ether (ISO) $\begin{matrix}CF_3\\CF_3\end{matrix}\!>\!CH\text{-}O\text{-}CH_3$ and observed it to possess anethetic activity comparable to ether in several species of animals.

We considered the two compounds ideally suited for detection of a physicochemical clue that might shed light on their respective activities. Pharmacologically, the responses evoked by these isomers are at the opposite ends of the spectrum of normal central system activity, namely, anesthesia and convulsions.

Materials and methods. The flurothyl was supplied by the Ohio Chemical and Surgical Equipment Co., Madison, Wisc. The isomer was supplied by Dr. Louise Speers, Air Reduction Co., Murray Hill, N. J. ISO is a clear, colorless liquid with an ethereal odor; B.P. 50° and Sp.G. 1.36.

The antagonism of the two isomers was studied in mice by the inhalation method of Cascorbi and Rudo(3).

The physicochemical methods were the well known standard procedures.

* Supported by a Grant from Life Sciences Division, Army Research Office.

TABLE I. Effect of ISO on Convulsive Properties of Flurothyl in Mice.*

Vol % ISO†	Vol % Flurothyl	No. convulsions	Time of onset of convulsions (sec)	No. dead
0	.25	8	30	1
.63	.25	1	300	0
1.25	.25	0	—	0
0	.50	10	5	9
.63	.50	9	40	0
1.25	.50	2	45	0
2.50	.50	0	—	0

* 10 mice in each group exposed for 10 min.
† Anesthetic concentration is 5.0 vol %.

Results. Table I shows that the addition of ISO in subanesthetic doses protects mice from flurothyl convulsions at 0.25 vol % and enables them to survive the lethal dose of 0.50 vol %. Further, it was observed that mice exposed to a mixture of 1.25 vol % ISO and 0.25 vol % flurothyl for 10 minutes did not exhibit any behavior characteristics that could be differentiated from normal mice (balanced state). The antagonism of the two isomers as shown in mice prevailed in rabbits, rats, and monkeys also.

An examination of the physical measurements in Table II reveals a difference in the dipole moments of ISO and flurothyl which are considered to have relevance in the potency of anesthetics. These do not appear to be capable of significant interpretation in view of the following data taken from the literature(4). For example, the dipole moments of other compounds are: $CHCl_3$ (anes-