nitude of change.

Bipolar registration, shifting of electrodes, use of needle electrodes or coupling of 2 to 8 electrodes did not yield any essential improvement. The changes in the records from the base of the skull were similar to those observed from the cortical area.

In 4 experiments with morphine an additional decrease occurred to some extent on the application of the pain stimulus. Further tests were not made because pain and morphine[2] tend to produce both flattening of the middle range of waves which makes reliable interpretation of experimental data practically impossible.

Summary. The possibility of E.E.G. registration of pain was examined in 17 human subjects. A decrease in amplitude of the waves was observed, which was more evident in the parietal, occipital, temporal and frontal in a descending order. This change, however, is not dependent on the nature and degree of pain and it is not specific as it occurred with cold, heat and touch as well.

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2 Gibbs, F. A., and Maltby, G. L., *J. Pharm. and Exp. Therap.*, 1943, **78**, 1.

Experimental Pulmonary Edema. II. Pathogenesis of Pulmonary Edema Caused by Ammonium Ion.* (17455)

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Koenig and Koenig[1] have described an acute pulmonary edema following the administration of ammonium salts to different species. It appears to be specifically a result of the ammonium ion. The occurrence and gross appearance of this edema resembles epinephrine induced pulmonary edema and in an effort to obtain information about the mechanism of the ammonium pathology a study has been made of its relation to edema due to epinephrine and adrenergic stimuli.

Experimental. Epinephrine induced pulmonary edema may be prevented by adrenergic blocking agents.[2,3] Their influence was observed upon ammonium pulmonary edema. Unfasted guinea pigs were divided into groups with the same sex distribution. All groups were lightly etherized to pass a stomach tube. Group 1, the controls, were given a sham dose of 10 cc water per kg body weight. All other groups received 1200 mg/kg of a 12% solution of NH_4Cl by oral gavage. Ten minutes before receiving the ammonium salt groups 3, 4 and 5 respectively received by intravenous injection in 1 cc saline/kg; 2 mg/kg of a B-haloethylamine, N-(9-fluorenyl)-N-ethyl-B-chloroethylamine-HCl ("SKF-501"); 2.5 mg/kg N-(2-chloroethyl)-N-ethylbenzhydrylamine-HCl ("SY-2"), which like "SKF-501" is an effective adrenergic blocking agent; and 1.5 mg/kg of the potent antihistaminic N-B-dimethylaminopropylthiodiphenylamine - HCl ("Phenergan", 3277 R.P.). The presence of pulmonary edema was determined at an autopsy by gross inspection and lung weight was used as a measure of its extent.

The summarized data comprise Table I. Group 1 was killed with ether while all of the animals in the other groups succumbed with convulsions 5-30 minutes after receiving the ammonium salt. The two adrenergic blocking agents and the antihistaminic all were

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1 Koenig, H., and Koenig, R., PROC. SOC. EXP. BIOL. AND MED., 1949, **70**, 375.

2 Stone, C. A., and Loew, E. R., PROC. SOC. EXP. BIOL. AND MED., 1949, **71**, 122.

3 MacKay, E. M., and Pecka, E. F., Jr., PROC. SOC. EXP. BIOL. AND MED., 1949, **71**, 669.

TABLE I.
Prevention of Ammonium Chloride Pulmonary Edema in Guinea Pigs with Adrenergic Blocking Agents.

Group	No. of animals: In group	No. of animals: With edema	Avg body wt, g	Lung wt, g/100 g body wt
		Controls		
1	18	0	736	0.69 ± .11
		1200 mg NH_4Cl/kg		
2	24	24	680	1.50 ± .21
	1200 mg NH_4Cl/kg preceded by 2 mg/kg of N-(9-fluorenyl)-N-ethyl-B-chlorethylamine*			
3	18	0	645	0.71 ± .11
	1200 mg NH_4Cl/kg preceded by 2.5 mg/kg N-(2-chloroethyl)-N-ethylbenzhydrylamine • HCl†			
4	12	0	685	0.65 ± .15
	1200 mg NH_4Cl/kg preceded by 1.5 mg/kg N-B-dimethylaminopropylthiodiphenylamine • HCl‡			
5	12	11	676	1.40 ± .25

* We are indebted to Dr. Glenn E. Ullyot, of Smith, Kline and French in Philadelphia for a supply of this compound (SKF-501).

† Dr. Leon A. Sweet of Parke, Davis and Co., Detroit, Mich., sent this material (SY-2) to us.

‡ (Phenergan, 3277 R.P.) supplied through the courtesy of Dr. A. Gibson, Assoc. Medical Director, Merck and Co., Rahway, N. J.

without influence upon mortality. Since both of the adrenergic blocking agents completely prevented the ammonium edema but not the mortality, we may conclude that pulmonary edema need not be the cause of death from toxic doses of ammonium salts. The anthistaminic was without effect on the ammonium edema, which in this respect resembles the fulminating pulmonary edema due to epinephrine.[2]

Epinephrine-induced pulmonary edema appears to be due to increased pulmonary pressure which it produces[2] possibly not directly, even though the vascular tree of the lungs is richly supplied with sympathetic fibres, but as a result of the high systemic arterial pressures. The complete protection against epinephrine edema afforded by adrenergic blocking drugs[2,3] which pharmacologically block excitatory actions of epinephrine such as vasoconstriction, is also true of the ammonium edema which suggests that the latter might also result from adrenergic stimuli. Some years ago we suggested[4] that the pulmonary edema of experimental hypoglycemia might be due to the release of epinephrine by the adrenal medulla. If this were true, medullectomy of the adrenal in the rat, where this procedure is feasible, could affect the development of the ammonium edema. We tried to test this but were unable to produce this type of pulmonary edema in our rats (Slonaker Strain[5]). Groups of 6 rats each had lung weights of 0.53 ± .06 g/100 g body weight (controls), 0.55 ± .07 g/100 g body weight (normals receiving 0.4 cc 10% NH_4Cl/100 g body weight intraperitoneally) and 0.48 ± .09 g/100 g body weight (demedullectomized rats receiving the same dose of NH_4Cl), respectively. This was equivalent to the dose of ammonium salt used by the Koenigs to produce edema in their rats. Nor could we produce pulmonary edema in the rat with ammonium salts whatever the dose when given *per os* as well as intraperitoneally. This is additional evidence that mortality from ammonium salts is not necessarily a result of the pulmonary edema which they frequently cause, since our rats all died. The same thing was true in albino mice of an unknown strain. We also are in disagreement with the Koenigs as to the action of ammonium salts in the rabbit. They found no edema. We regularly found lung edema after the administration of am-

[4] Sherrill, J. W., and MacKay, E. M., *Arch. Int. Med.*, 1939, **64**, 907.

[5] MacKay, L. L., and MacKay, E. M., *Am. J. Physiol.*, 1927, **83**, 179.

TABLE II.
Influence of Ammonium Chloride on Blood Sugar.

			Average blood sugar concentration—mg%*	
Time in min.	1 Controls	2 NH_4Cl	3† NH_4Cl preceded by "SKF-501"	4‡ NH_4Cl preceded by tetraethylammonium
0	112	132	140	136
10	116	216	224	206
17	118		208	
20			260	220
22		272	256	244
24	120	296	272	282

* 3 unfasted guinea pigs in each group.
† Doses of NH_4Cl and "SKF-501" as in Table I.
‡ Dose of tetraethylammonium bromide 10 mg/kg subcutaneously.

monium chloride and it was prevented by the adrenergic blocking agents. The control lungs always appeared much more edematous than their weight would indicate. We have often noted a disproportion between the extent of edema and the amount of fluid which collected in the alveoli and bronchioles. Groups of four 2 kg rabbits were given 1.25 g/kg of 25% NH_4Cl by stomach tube. Two groups were given the blocking agents in the same dose used in the guinea pig experiments. The lungs of Group 1 (controls) averaged 0.46 g, Group 2 (NH_4Cl treated) 0.71 g, Group 3 (NH_4Cl preceded by "SKF-501", see Group 3, Table I) 0.45 g and Group 4 (NH_4Cl preceded by "SY-2", see Group 4, Table I) 0.42 g/100 g body weight, respectively. There was no edema except in the lungs of Group 2.

Ammonium salts might exert an adrenergic effect by direct action on sympathetic nerve endings, or by direct or reflex central stimulation. Ammonia has long been known as a most effective means of stimulating the brain medulla reflexly[6] and in addition to lung edema, the Koenigs have found pathology resulting from fatal doses of ammonium salts. Hypnotics have been claimed to prevent lung edema due to epinephrine.[7] Ethyl ether produces an anesthesia characterized by complete paralysis of the motor reflex centers in the cord and some depression of the medullary centers. When our animals were continued under a deep ether anesthesia in place of stopping the anesthetic after the light etherization used for gavage to give the ammonium chloride, they lived as long as similar unanesthetized controls and died without the usual convulsions typical of toxic doses of ammonium salts. Pulmonary edema was always absent. The average lung weight of 4 etherized ammonium chloride pigs was 0.57 in comparison with 1.34 g/100 g body weight for a similar group of non-etherized NH_4Cl controls. We interpret this as evidence in favor of the ammonium-induced pulmonary edema being a result of reflex or direct stimulation of the medullary centers which results in an outflow of excessive adrenergic stimuli. The stimuli do not necessarily act on the pulmonary circulation primarily but as in epinephrine pulmonary edema[3] may produce left heart failure with an increased pulmonary venous pressure and the inevitable edema.

The administration of ammonium chloride causes a marked hyperglycemia which might be interpreted as due to a release of epinephrine by the adrenal medulla. However ammonium salts have been reported[8,9] to have no effect on adrenal release of epinephrine and they may induce a hyperglycemia in adrenalectomized rabbits.[10] We have found (Table II) that neither an adrenergic blocking agent nor tetraethlammonium which blocks the release of epinephrine by the adrenal medulla[11] influences the hyperglycemia due to ammoni-

[6] Sollman, T., A Manual of Pharmacology, 3rd ed., p. 859, W. B. Saunders Co., Philadelphia, 1928.
[7] Luisada, A., *Arch. Exp. Path.*, 1928, **132**, 313.
[8] Houssay, B. A., and Molinelli, E. A., *Compt. rend. Soc. biol.*, 1925, **93**, 1156.
[9] Shoji, J., *Tohoku J. Exp. Med.*, 1937, **30**, 259.
[10] Satake, Y., *Tohoku J. Exp. Med.*, 1926, **8**, 26.

um salts in guinea pigs and conclude that the carbohydrate disturbance is not directly related to the mechanism of the pulmonary edema.

Summary 1. The acute pulmonary edema which follows the administration of ammonium salts to guinea pigs may be entirely prevented by prior administration of the adrenergic blocking agents, N-(9-fluorenyl)-N-ethyl-B-chloroethylamine-HCl ("SKF-501") and N-(2-chloroethyl)-N-ethyl-benzhydrylamine-HCl ("SY-2"). Less marked lung edema is produced in rabbits by ammonium chloride and it is also prevented by the blocking agents.

2. A potent antihistaminic agent, N-B-dimethylamino-propylthiodiphenylamine-HCl ("Phenergan-3277 R.P."), was without effect on the incidence of ammonium pulmonary edema, which in this respect as well as the manner it is influenced by adrenergic blocking agents, resembles epinephrine-induced pulmonary edema.

3. A central nervous system depressant in the form of continuous ether anesthesia prevents the occurrence of ammonium pulmonary edema. Along with the effect of adrenergic blocking agents this is construed as evidence for the mechanism of this edema being due to adrenergic stimuli of reflex or direct central stimulation origin.

4. Ammonium salts cause a striking hyperglycemia. Since adrenergic blocking agents of the type used as well as tetraethylammonium which blocks epinephrine release by the adrenal medulla fail to prevent the hyperglycemia response it is believed that ammonium hyperglycemia is not directly related to the edema formation.

5. Although pulmonary edema sometimes may be the cause of death from toxic doses of ammonium salts, this is not true in the Slonaker strain of rats or our strain of albino mice both of which fail to develop edema. Nor is this the cause of death in guinea pigs or rabbits when an adrenergic blocking agent is used to prevent the edema, as the animals succumb as a result of other effects of the ammonium ion.

Addendum: When this manuscript was ready for submission to the Editors, an article by the Koenigs (Koenig, H., and Koenig, R., *Am. J. Physiol.*, 1949, **158**, 1) on the pathogenesis of ammonium pulmonary edema appeared. Insofar as our observations are similar the conclusions are parallel.

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11 Morrison, J. L., and Farrar, C. H., PROC. SOC. EXP. BIOL. AND MED., 1949, **71**, 235.

Effect of Adrenal Cortical Extract and of X-Radiation on Serum Proteins of Mice. (17456)

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It has been reported previously from this laboratory that chronic injections of pituitary adrenotrophic hormone in mice produced an increase in the total serum proteins.[1] Moreover, a single injection of adrenotrophin or of adrenal cortical steroids in oil in rabbits was observed to cause an elevation of circulating β- and γ-globulins within a few hours after hormone administration.[2] Electrophoretic analysis also revealed an increase in these

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1 White, A., and Dougherty, T. F., PROC. SOC. EXP. BIOL. AND MED., 1944, **56**, 26.

2 White, A., and Dougherty, T. F., *Endocrinology*, 1945, **36**, 207.