TOXICITY OF INTRAVENOUS AMMONIUM SALTS

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A THESIS

Presented to the Department of Pharmacology
and the Graduate Division of the University of Oregon Medical School
In partial fulfillment of the requirements for
the degree of Master of Science

June 1948

APPROVED:

[redacted]

(Professor in Charge of Thesis)

[redacted]

Chairman, Graduate Council

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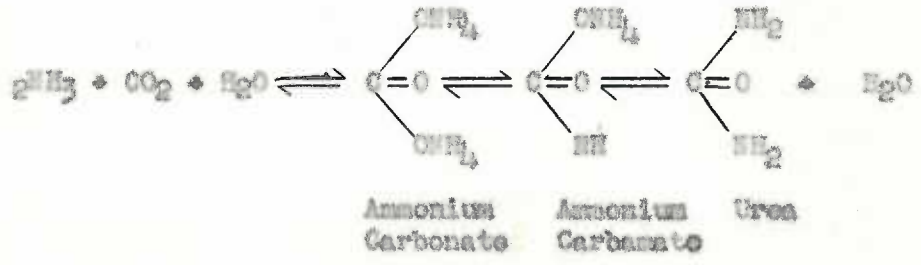
INTRODUCTION

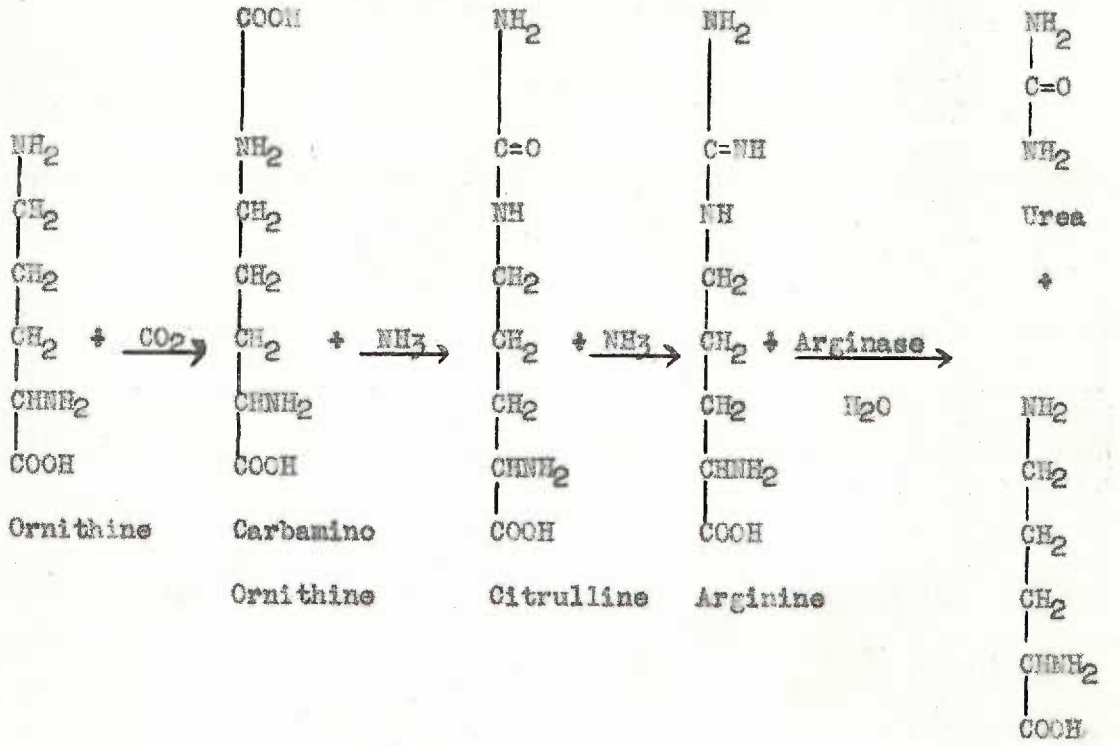
1 Chemistry and Metabolism of Ammonium Salts.-

The position of the ammonium ion in physiological chemistry is unique because it is the only inorganic cation which may be synthesised by a body organ into a nontoxic organic compound. This organ is the liver and its efficiency, in this respect, is so great that the normal ammonia content in the circulating human blood is constantly maintained at, or very near zero (3). Urea is the organic compound to which ammonia is converted and the mechanism is believed to take place by two procedures: The first is the simple combination, probably through enzymatic action, of ammonia, carbon dioxide and water to form first ammonium carbonate, then ammonium carbamate and, finally, urea and water. In the other process, the amino acid ornithine is carboxylated by the addition of carbon dioxide to carbamino ornithine. Addition of NH_3 to this later compound changes it to citrulline, which, with the addition of further ammonia changes it to the amino acid arginine. The action of arginase on the arginine then releases urea and provides ornithine again to allow this interesting organic synthetic process to begin over again. The following formulae (1) and (2) show these two separate steps which maintain the constancy of the zero or near zero blood level of ammonia:

1.



2.



When ammonium chloride is ingested or injected, after the metabolism of the ammonium with conversion to urea, the chloride ion remains free in the circulation. In turn, this cation represents a metabolic problem since it must be neutralized by an equivalent ion of fixed base (Na ion) in order to maintain acid-base equilibrium for homeostasis.

For clarity a more detailed explanation fo this mechanism is that while the ammonium ion is converted to urea, the liberated chloride ion draws upon sodium ions contributed mainly from the sodium bicarbonate buffer system of the plasma. This leads to a formation of an excess of carbonic acid which is quickly excreted through the lungs as carbon dioxide, and results in a state of compensated alkali deficit with reduced plasma carbonic acid and sodium bicarbonate although the pH remains normal. Since the carbon dioxide combining power is a measure of the amount

of plasma bicarbonate, consequently it is lowered. If the amount of chloride ion present is very great, the plasma sodium bicarbonate becomes severely depleted. The carbonic acid (HCO_3-) released from the $NaHCO_3$ combination accumulates since the lungs are unable to excrete the freed CO_2 rapidly enough and a fall in the plasma pH, as well as the CO_2 combining power occurs. Thus a state of uncompensated alkali deficit and CO_2 excess now exists.

The kidney is also intimately connected with the metabolism of the ammonium ion. It serves as the only avenue of excretion for the increased amount of urea and sodium chloride formed by the metabolism of the ammonium chloride in the liver. Of some importance is the fact that with a scarcity of fixed base, the cells of the renal tubules can form ammonia from certain amino acids (Krebs) or even urea itself (Mann and Bollman, Nash and Benedict) (1). This ammonia of renal origin displaces the sodium combined with acid anion thus allowing the tubules to reabsorb the sodium and so preserving this cation to the body. Under these circumstances the kidney also selectively excretes a great amount of NaH_2PO_4.

In addition to its effects on acid-base balance, ammonium chloride indirectly exerts an influence on the body water balance. Both urea and sodium chloride have considerable osmotic activity, and consequently, when these compounds are being excreted in large amounts, a large amount of water accompanies them because tubular reabsorbtion of water from glomerular filtrate is decreased by the high osmotic pressure of these compounds.

2. Review of Literature on Experimental and Clinical Studies with Administration of Ammonium Salts.-

In 1921, Haldane (12) first demonstrated the ability of ammonium chloride to lower the blood carbon dioxide combining power indicating a shift toward the acid side after the oral ingestion of large quantities of this compound. It is obvious from the effects mentioned that ammonium chloride would be of value therapeutically. The drug has long been administered orally, primarily as a diuretic either alone or in conjunction with digitalis and the mercurial diuretics.

Less frequently, intravenous administration of ammonium chloride has been practiced. McCann (15) in 1922 reported on the relief of carpopedal spasm from gastric tetany and alkalosis by the intravenous injection of 500 ml. of 0.822 per cent ammonium chloride solution. In 1925 Youmans and Greene (23) also reported on the successful treatment of a case of gastric tetany by the intravenous administration of 400 cc of 0.82 per cent ammonium chloride and observed no untoward reactions, but, from 1925 until 1943, this form of therapy for alkalosis was supplanted by the intravenous administration of large quantities of physiologic salt solution (0.9% NaCl in distilled water), mainly because of the work of Gamble and Ross (10. These authors stated that if the kidney were presented with adequate amount of sodium and chloride ions, there would take place a selective excretion of the anion (Cl^-) with preservation of the cation Na^+) thus reestablishing the acid-base equilibrium. They also pointed out that intravenous ammonium chloride may be harmful in the treatment of alkalosis because it may cause depletion of sodium ion and aggravation of dehydration by diuresis.

However, Nicol (17) in 1940, pointed out that in alkalosis some renal damage or insufficiency may occur so that the kidney is unable to function in its normal physiologic manner. In addition to renal damage, severe alkalosis may produce tetany, convulsions, and death. Thus, in 1943, the use of ammonium chloride was resumed by Sintel, Rhoades, and Rawdin (24) who reported on the treatment of seven patients in alkalosis who had received a total of twelve intravenous injections of 2% ammonium chloride contained in either distilled water, physiologic saline, or 5% glucose solution.

In 1945, Sellers and Kast (19) described the use of intravenous ammonium chloride in three patients. In each case its injection lead to a prompt fall in carbon dioxide combining power and a rise in serum chlorides. Sellers and Kast in contrast with Gamble and Ross (9) suggested that many patients fail to respond to therapy with physiologic salt solution because of the impaired renal function due to alkalosis, nephrosis, dehydration, hypochloremia, and/or concommittant renal disease. In this condition nausea, vomiting, coma and tetany preclude oral administration and makes rapid treatment desirable.

In 1946, Forbes and Ergenian (8) administered intravenous ammonium chloride to nine infants with congenital hypertrophic pyloric stenosis following an experimental study of its effects on dogs and rabbits. They employed a 0.89% solution of ammonium chloride (one-sixth molar) in either isotonic sodium chloride or in isotonic solution of three chlorides U.S.P. (Ringer's solution). The rational employing sodium chloride to replace the loss, in vomitus, of an appreciable amount of sodium ions had been pointed out by Gamble and Ross (10). Forbes and Ergenian found that the intravenous and subcutaneous route of administration of ammonium

chloride was effective in lowering the carbon dioxide combining power of the plasma and raising the serum chlorides. In animals, they found that the intravenous injection of 20 to 30 cc per kilogram body weight in 45 minutes (equal to .074 to 0.111 milliequivalents per kilogram per minute) caused symptoms of excessive salivation, hyperpnea, and drowsiness. Increasing the rate of injection caused stupor and convulsions and it was noted that the symptoms produced were directly related to the rate of injection of the solution. During clinical use one infant showed definite toxic effects with pallor, irregular respiration, occasional twitchings of the eyelids and hands, bradycardia, and poor response to painful stimuli. This child had been given 30 cc per kg. of 0.89% solution of ammonium chloride in eighteen minutes (0.276 milliequivalents per kilogram per minute). The authors concluded that the toxicity was due to too rapid injection of the solution, and a concomittant production of an extensive fall in serum pH.

Schoen (1947) (18) used a 0.46% solution of ammonium chloride in 2% dextrose solution by vein as a diuretic in treating resistant edema and oliguria. Five of his cases, totaling 52 administrations, showed severe reactions characterized by pallor, sweating, and retching.

For a compound such as ammonium chloride which gives promise of considerable therapeutic use, it is important that a thorough qualitative and quantitative knowledge of its pharmacology be gained. The qualitative aspects attending intravenous injection of ammonium chloride have been well known for many years. Following large intravenous doses of ammonium chloride, Marfori (20) in 1893, described the prompt appearance of twitches, tremors, progressing to tetany, violent convulsions, and opisthotonos, irregular respiration, salivation and emesis,

and somnolence and lassitude. Meneguissi (20) (1912) stated that the body functions recover rapidly providing the injection is not immediately fatal. Brassfield (20) et al (1946) found that intravenous injection of ammonium chloride produced an immediate increase in respiration, fall in blood pressure, and slowing of the heart. Some of the other reported actions of ammonium chloride include stimulation of the central nervous system with toxic amounts, a weak curare-like action on direct application to muscle, and an expectorant action.

With respect to the quantitative aspects of the toxicology of ammonium chloride, these are less well known. The above reports, especially those of Meneguissi, and Forbes and Ergarian would suggest that the occurrence of toxicity is related more closely to rate of administration than to total amount administered. However, an extensive search of the literature has failed to disclose any studies primarily concerned with this particular question.

METHOD OF STUDY

Our purpose in this investigation has been (1) to determine the nature and sequence of toxic signs, (2) to quantitatively determine the rate or rates of injection which will produce toxicity, and (3) to determine the rate of injection which will produce death.

1. Injection of Ammonium Salts.-

Healthy, fasting (24 hrs.), adult, male and female dogs anesthetized with sodium pentobarbital (35 mg./kg.) were used. The intravenous infusions were made in 5% dextrose solution as either 2% ammonium chloride, 2.88% ammonium acetate, 2.96% ammonium bicarbonate, 2.0% or 2.5% ammonium carbonate. A 2% solution of ammonium chloride was chosen because

this was the solution used clinically by Zintel, Rhoades, and Rawdin (16) and Sellers and East (17). The percentages of the other solutions were chosen because they are equivalent in ammonium ion concentration to the 2% ammonium chloride. The rate of infusion was determined by frequently counting the drops falling in 30 seconds time in the intravenous apparatus. At the end of the experiment the number of drops comprising 10 ml. of the particular salt used was determined from an average of three different readings. This was interpolated into the number of drops per milliliter. For the sake of comparison the rates were converted to milliequivalents of ammonium ion per minute and finally to milliequivalents per kilogram body weight per minute.

2. Determination of Blood Ammonia.-

Blood ammonia was determined by the method of Conway (2, 3, 4, 5) but since our results vary from his, the method we used is described in detail. The "units" used for the reaction, round cups 60 mm. in diameter and 10 mm. deep and having an inner and outer chamber, were the same size as those described by Conway but were made of plastic instead of pyrex glass. Preliminary preparation of the "units" was that recommended by Conway, namely: the center chamber was filled with 0.7 ml. of 0.0002 N sulfuric acid and the outer chamber with 1.0 ml. of saturated potassium carbonate, the lid smeared with mineral oil-paraffin fixative and set in place. This procedure was done to remove traces of ammonia in either the fixative, the unit, or the potassium carbonate. At the end of one-half hour or longer the acid in the central chamber was titrated to the end-point with 0.0005 N barium hydroxide, then withdrawn, and a refill of acid was put in the center

chamber. Conway collected blood either by open shedding thru a needle connected to a short length of paraffined rubber tubing into an open flask, or by aspirating it into an atmosphere of carbon dioxide in a tonometer. Our procedure was to collect the blood into a syringe containing mineral oil thru a 21 or 22 gauge needle and transferring it under a layer of mineral oil into a test tube. A stop watch was started at the moment blood first appeared in the syringe and stopped at the end of the rotation of the "unit" in order to calculate the ammonia formed during the alpha rise of Conway which will be discussed later. The "unit" was then tipped and the lid removed enough to admit the tip of the pipette. One milliliter of blood was transferred to the outer chamber by means of a Van Slyke-Neill pipette and the lid quickly replaced. The unit was rotated ten times to mix the blood with the potassium carbonate and set aside for exactly ten minutes. The excess acid was then titrated with barium hydroxide. In the first few determinations, the ammonia content was calculated on the basis of the normality of the acid and base, but in later experiments, control and blank estimations were run frequently and the following formula used to calculate the micrograms of ammonia per milliliter of blood. The value of this method of calculation is that the burette and reagents are standardised against a control solution of known strength.

$$\frac{\text{(Blank minus analytical}}{\text{Blank minus control}} \times 1.13 \times 2.65) - 0.11 = \text{ugm } NH_3 \text{ nitrogen per ml.}$$

Where: "Blank" is the amount of base, read as marks on the burette, necessary to neutralise the acid after a determination with distilled water. Analytical equals marks on the burette to neutralise the acid after ammonia from 1 ml. of blood has been absorbed by the acid. Control

equals marks on the burette to neutralise the acid after absorption of ammonia from 1 ml. of a control solution containing 2.65 ugm NH_3 nitrogen per ml. 1.15 is the ratio of NH_3 absorbed from water carbonate mixtures to that from blood carbonate mixtures (Conway, 5). 2.65 is the concentration of ammonia nitrogen in control, in ugm per ml. The figure 0.11 is the correction for special deaminating action of the alkali on blood at 25° C. in ugm of ammonia nitrogen per ml. (Conway, 5).

The same procedure was followed with blood of higher ammonia content which is found during infusion of ammonium chloride, except that an acid of higher normality is used. The same blank and control estimations were done.

Conway (3) first showed in 1935 that the normal human blood ammonia content is practically zero. A rapid rise in ammonia occurs in blood which is collected by open shedding, reaching 0.4 to 0.5 ugm ammonia nitrogen per ml. by the end of 5 minutes. This has been called the "alpha" rise by Conway and is presumably due to deamination of minute amounts of adenosine and adenylic acid. A more gradual increase occurs after this time, called the beta rise, and a third, still slower rise called the gamma phase, occurs later.

Kaprowski and Uhinski (13) showed that the normal blood ammonia in dogs is also zero, and that the same alpha rise occurs as in man. They also showed that the blood ammonia increases after the oral administrations of ammonium chloride (0.5 gm. per kg.), the highest value being 10 ug per ml. Blood urea also rises, the steepest rise occurring from 60-90 minutes after administration of ammonium chloride.

In our studies, the carbon dioxide combining power of the plasma was determined by the Van Slyke manometric method.

EXPERIMENTAL RESULTS

A. Typical toxic signs observed from intravenous injection of ammonium salts.

We have observed in the majority of cases three characteristic stages of reaction in intravenous ammonium chloride. These may be divided into the beginning, the train, and the final toxic signs. In 26 dogs and with the use of any of the four ammonium salts, the typical series of events was as follows:- First, there were regularly occurring deep inspiratory gasps followed by a marked expiratory effort which was produced by a strong contraction of the abdominal muscles. These became increasingly more frequent and soon the respirations became quite irregular. Recording of the carotid blood pressure showed that at the same time the systolic pressure began to rise and the stroke volume became very great. Soon after this occasional muscular twitches appeared, usually beginning about the neck, angles of the mouth, eyelids, and lips. There were fasciculations of the muscles of the tongue. By this time the respirations had become completely irregular and were often accompanied by a violent gasping inspiratory effort with a marked ventral jerking of the head. The muscular twitchings spread throughout the body and were soon followed by tonic and clonic convulsions. Unless the injection was stopped, death ensued from respiratory arrest, for the heart continued to beat for a few seconds after the respirations ceased. Other toxic effects as reported by others were noted, such as cardiac irregularities, auscultatory sounds characteristic of fluid in the lungs (14), and marked

hemolysis of the red blood cells.

In 22 of the experiments the infusion was allowed to continue until the animal was dead. Death occurred on an average of 31.25 minutes after the toxic symptoms first appeared with extremes at 11 and 70 minutes. In 11 animals the intravenous infusion was stopped soon after the onset of toxicity and the dog allowed to recover. The average period necessary for recovery was 28 minutes with extremes at 8 and 65 minutes. The time for recovery to occur was directly proportional to the length of time toxicity had been allowed to exist.

B. Atypical Toxicity.-

In 4 experiments the typical train of toxic events did not occur. In these cases, the rate of infusion was fast. In one instance, the first thing noted was a tonic convulsion followed immediately by death. In the second case, there was some trembling noted at the end of inspiration and the animal quietly ceased breathing. In the third animal, there was irregularity of the respirations but no fibrillations or convulsions and the animal died by respiratory arrest without other warning signs. The fourth dog had several gasping respirations immediately followed by two tonic convulsions, vomited and died from respiratory failure.

Following are abstracted protocols giving the train of events in four typical experiments, exemplifying the toxicity of ammonium chloride, ammonium carbonate, ammonium bicarbonate, and ammonium acetate.

1. 2% NH_4Cl 12.9 kg. Female Dog - Sodium Pentobarbital Anesthesia.

TIME (Min.)	RATES of INFUSION (meq./kg. minutes)	CONDITION
0	-	Normal.
14	0.222	Convulsive efforts during inspiration, jerking of head, spasm of groups of muscles, lungs full of fluid.
25	0.208	Heart slow and irregular, twitching of head, neck, legs, eyes, and eyelids.
32	0.201	Two successive tonic and clonic convulsions.
41	0.194	Fasiculation of the tongue, marked inspiratory gasps.
43	0.208	Clonic convulsion, death from respiratory failure.

2. 2.96% NH_4HCO_3 10.9 kg. Male Dog - Sodium Pentobarbital Anesthesia.

TIME (Min.)	RATES of INFUSION (meq./kg./minutes)	CONDITION
0	-	Normal
11	0.169	Pulse pressure high, slightly irregular pulse, blood pressure high, moderate cyanosis.
17	0.169	Twitching of neck muscles with inspiration, twitching of abdominal muscles, pulse irregular.
36	0.155	Jerking of head with inspiration, heart rhythm erratic, urinary incontinence.
44	0.129	Tonic convulsion, very irregular and gasping respirations, fibrillary twitching of tongue.
69	0.132	Tonic convulsion, urinary incontinence.
70	0.132	Death from respiratory failure.

3. 2.08% NH_4Ac 10.5 kg. Female Dog - Sodium Pentobarbital Anesthesia.

TIME (min.)	RATES of INFUSION (meq./kg./min.)	CONDITION
0	-	Normal - P.212 and regular, R26 and regular, color good, reflexes sluggish.
11	0.196	Forceful abdominal contraction with expiration.
32	0.196	Fibrillary twitchings of the tongue.
35	0.197	Sinus arrhythmia, respiration irregular, uneven depth, jerky, slight cyanosis.
42	0.196	Wink reflex now present, swallowing.
50	0.197	Generalized muscular twitching, gasping respirations, cyanosis, jerking of head, jerky irregular spasmodic contractions of abdomen between breaths, sinus arrhythmia.
60	0.191	Marked fasciculation of tongue, tonic convulsion.
62	0.191	Death from respiratory failure.

4. 2.0% $(NH_4)_2CO_3$ 10.8 kg. Female Dog - Sodium Pentobarbital Anesthesia.

TIME (min.)	RATES OF INFUSION (meq./kg./min.)	CONDITION
0	-	Normal
11	0.155	Respirations 20/min., and regular. Pulse 94/min., and regular.
35	0.159	Gasping respiration, fasciculations of tongue, quivering, convulsive movements. IV slowed and dog allowed to recover.
48	0.029	Gasping respiration, no tremors or fasciculation. Pulse rate 140/min., and regular, moderately strong.
75	0.026	Pulse rate 140/min., and regular, moderately strong. Respiration 25/min. and regular. Forcible expiration, color normal, wink and corneal reflexes absent, muscle tone moderate. Tendon reflexes hyperactive.

Table I summarized the data for the time of onset of toxicity in 30 dogs. The columns, reading from left to right show the weight of the dog in kilograms, the average rate of infusion of the ammonium salt in milliliters per minute, the conversion of the figures given for the average rate to millequivalents of ammonium ion per minute, the average rate of infusion in milliequivalents per kilogram per minute obtained from the quotient of the two previous figures, and the length of time after starting the intravenous infusion required for the appearance of toxic signs, the last column shows Re/Rc or the average experimental rate of infusion in milliequivalents per kilogram per minute (Re) divided by 0.045 (Rc) which is the rate of infusion in milliequivalents per kilogram per minute recalculated from the clinical program used by Zintel, Rhoades, and Ravdin (24) and Sellers and Kast (19). From our studies we found infusion as little as 2.3 times the recommended rate is capable of producing toxicity in 63 minutes and a rate of 6.48 times the recommended rate produced toxicity in 8 minutes.

In Graph I is shown the time of the onset of toxicity of all 30 dogs with the average rate of infusion in milliequivalents per kilogram per minute plotted against time in minutes.

Table II shows the time for death after the onset of toxicity at various average rates of infusion. Column 1, is the weight of the dog, column 2, is the average rate of infusion in milliliters per minute, column 3, is the average rate of infusion in milliequivalents per minute, column 4, is the dose in milliequivalents per kilogram per minute, and column 5 the time in minutes for death after the onset of toxicity for 20 dogs. From the table it is evident that death occurred immediately after the onset of toxicity in 2 dogs, and that another

TABLE I

INTRAVENOUS INJECTION OF VARIOUS AMMONIUM SALTS IN 5% GLUCOSE SOLUTION GIVEN TO DOGS

AMMONIUM SALT GIVEN	WEIGHT OF DOG (kg.)	RATE OF ADMINISTRATION ml./min. (aver.)	RATE OF ADMINISTRATION meq./min. (aver.)	DOSE meq./kg./min.	ONSET OF TOXIC SIGNS (mins.)	Ke/Ko
2% NH_4Cl	14.1	3.88	1.45	0.103	63	2.29
	10.9	3.39	1.27	0.116	63	2.50
	21.4	7.67	2.86	0.133	45	2.69
	10.9	3.61	1.35	0.124	34	2.76
	6.4	3.19	1.19	0.143	64	3.18
	7.3	2.93	1.10	0.151	32	3.36
	17.7	8.43	3.15	0.178	17	3.96
	7.7	3.98	1.49	0.193	42	4.29
	8.2	4.25	1.59	0.194	36	4.30
	10.9	5.83	2.17	0.199	31*	4.42
	12.5	7.33	2.74	0.219	18	4.87
	12.9	7.69	2.88	0.223	14	4.96
	7.2	4.67	1.74	0.242	11	5.37
	6.7	5.26	1.97	0.292	8	6.48
NH_4Cl used clinically in man.	70.0	8.34	3.12	0.045	60	
2.00% AMMONIUM ACETATE	10.5	5.52	2.06	0.196	50	4.35
	11.4	6.06	2.27	0.199	39	4.42
2.0-2.5% AMMONIUM CARBONATE	13.3**	3.84	1.68	0.127	57	2.82
	10.8***	4.90	1.71	0.159	38	3.54

* Dog dead at that time

** 2.5%

*** 2.0%

TABLE I

INTRAVENOUS INJECTION OF VARIOUS AMMONIUM SALTS IN 5%

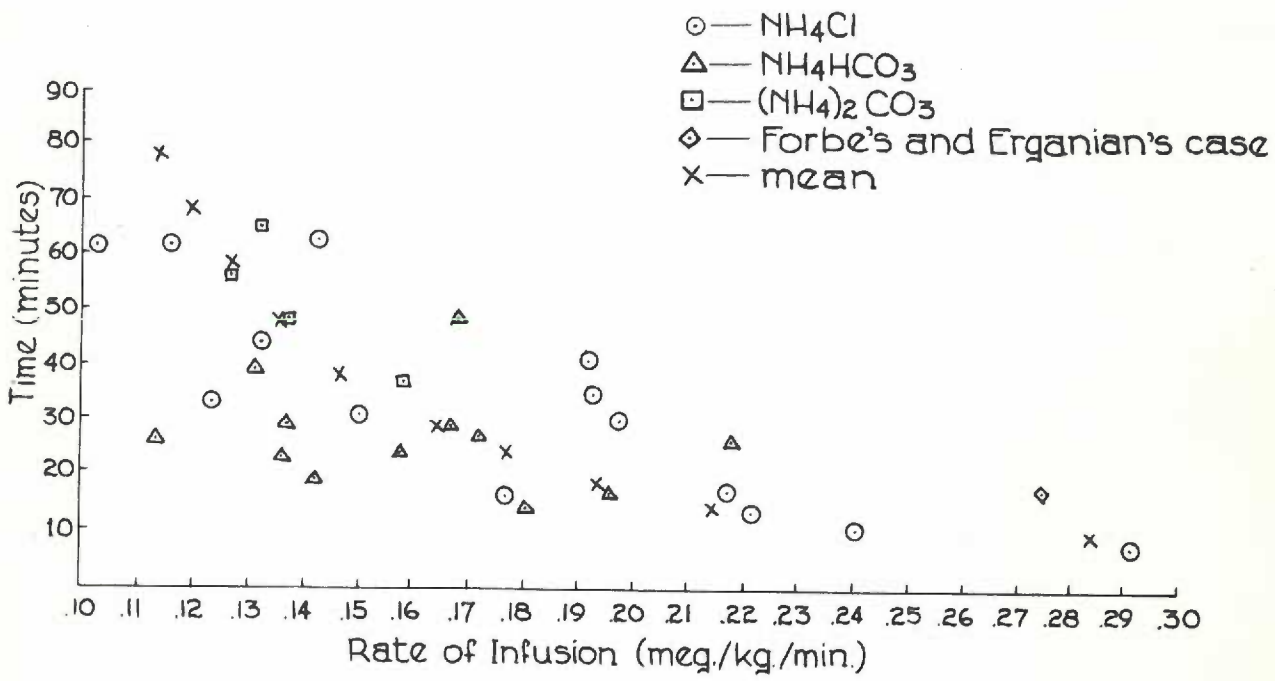
GLUCOSE SOLUTION GIVEN TO DOGS

AMMONIUM SALT GIVEN	WEIGHT OF DOG (kg.)	RATE OF ADMINISTRATION ml./min. (aver.)	RATE OF ADMINISTRATION meq./min. (aver.)	DOSE meq./kg./min.	ONSET OF TOXIC SIGNS (mins.)	Re/Ro
2.96% AMMONIUM BICARBONATE	10.8	3.30	1.23	0.114	27	2.53
	15.0	5.30	1.98	0.132	40	2.94
	13.6	4.87	1.82	0.133	66	2.95
	6.3	2.28	0.85	0.137	50	3.03
	10.0	3.66	1.37	0.137	24	3.04
	10.9	4.05	1.52	0.139	30	3.06
	7.2	2.79	1.03	0.143	20*	3.17
	13.3	5.65	2.12	0.159	25	3.54
	13.6	6.13	2.29	0.168	30	3.73
	13.3	6.00	2.24	0.169	50	3.77
	5.9	2.74	1.03	0.173	28	3.82
	6.8	3.30	1.23	0.182	15	4.05
	10.9	5.72	2.14	0.197	16	4.37
	8.9	5.21	1.95	0.219	27	4.87

* Dog died at that time.

GRAPH I

Relationship of time
to rate of infusion
for the onset of toxicity.



dog died 5 minutes after onset of toxicity although he may have aspirated vomitus. In the remaining 17 dogs, 11 minutes was the shortest time and 70 minutes was the longest time from the onset of toxicity to death.

Graph II shows the relationship between the average rate of infusion in milliequivalents per kilogram per minute and the time in minutes for the occurrence of death after onset of toxicity.

Table III lists the blood NH_3-N and CO_2 combining power before the start of the experiment (normal), at the onset of toxicity, and at death.

It will be seen that the plasma CO_2 combining power was lowered in all instances when ammonium chloride was used. The injection of neither ammonium bicarbonate nor ammonium acetate apparently had any marked effect on the plasma CO_2 combining power. In this respect, it is important to note also that no dogs showed a plasma CO_2 combining power in the range of alkalosis because of the similarity in the manifestations of alkalosis and those of ammonium ion toxicity. Also of note is the fact that there was no appreciable difference in time for the onset of toxicity or death with ammonium chloride or the other ammonium salts. Thus the toxicity and death is apparently due to the ammonium ion itself and not to changes in acid-base balance, as suggested by Forbes and Organism.

Blood ammonia levels at the appearance of toxic signs at death are quite variable and perhaps inconclusive because of the small number of determinations and also because we were unable to exactly reproduce the normal levels found by Conway (2, 3), and Kaprowski and Uninski (13). It is important to show as has been done in our

TABLE II

THE OCCURRENCE OF DEATH AFTER THE ONSET OF TOXICITY

AMMONIUM SALT GIVEN	WEIGHT OF DOG (kg.)	RATE OF ADMINISTRATION ml./min. (aver.)	RATE OF ADMINISTRATION meq./min. (aver.)	DOSE meq./kg./min.	TIME OF DEATH FROM ONSET OF TOXIC SIGNS (mins.)	Ro/Ro
2.0-2.5% AMMONIUM CHLORIDE	21.4	7.67	2.86	0.133	30	2.96
	7.3	2.95	1.09	0.149	70	3.31
	17.7	7.11	2.65	0.150	20	3.33
	8.2	3.31	1.23	0.151	29	3.36
	7.7	3.65	1.36	0.169	30	3.78
	6.7	5.00	1.89	0.283	12	4.20
	12.9	6.61	2.47	0.191	30	4.24
	12.5	6.61	2.47	0.197	18	4.38
	10.9	5.83	2.17	0.199	0*	4.42
	7.2	4.67	1.74	0.242	11	5.38
	6.4	4.39	1.64	0.256	15	5.69
2.9% AMMONIUM BICARBONATE.	10.0	3.28	1.23	0.123	44	2.73
	15.0	5.15	1.92	0.128	65	2.84
	10.9	3.89	1.45	0.131	40	2.91
	7.2	2.79	1.03	0.143	0*	3.18
	13.6	6.12	2.28	0.168	13	3.73
	13.3	5.65	2.12	0.169	5**	3.75
	10.9	5.72	2.14	0.197	24	4.38
	5.9	3.48	1.30	0.220	20	4.89
	0.9	5.42	2.01	0.226	22	5.02

* Died without previous warning signs.

* Died without previous warning signs.
** Vomited with possible aspiration.

GRAPH II

Relationship of time to rate of infusion for the occurrence of death after the onset of toxicity

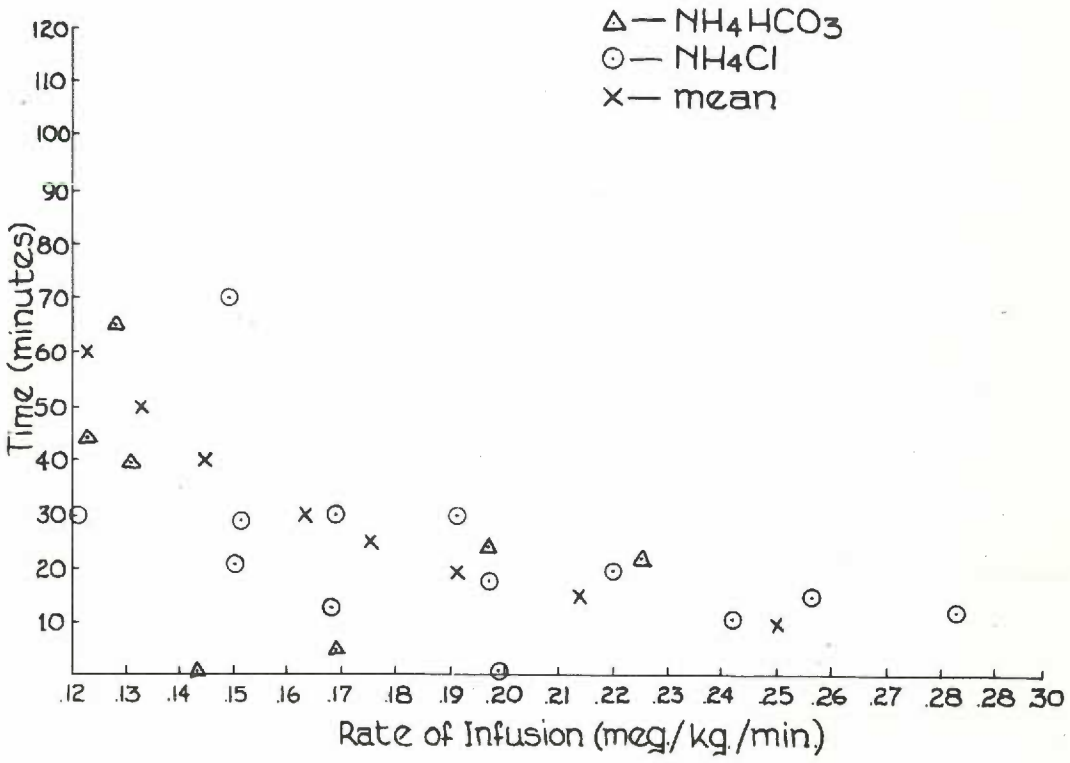


TABLE III

BLOOD AMMONIA-NITROGEN AND CARBON DIOXIDE COMBINING POWER

DRUG	NORMAL NH_3-N µg./ml.	NORMAL CO_2 COMBINING POWER (vol.%)	AT ONSET OF TOXICITY NH_3-N µg./ml.	AT ONSET OF TOXICITY CO_2 COMBINING POWER (vol.%)	AT DEATH NH_3 µg./ml.	AT DEATH CO_2 COMBINING POWER (vol.%)
NH_4Cl	0.42	56	-	-	39.1	40
NH_4Cl	0.40	52.5	-	48.5	61.7	11
NH_4HCO_3	1.25	90	-	-	52.7	63
NH_4HCO_3	1.16	53	43.8	61	45.2	53.5
NH_4Ac	1.13	53	21.3	45	45.4	46.5
NH_4HCO_3	-	-	-	44	-	56
NH_4Cl	-	40.5	-	24.5	-	18
NH_4HCO_3	-	39	-	53	-	46
NH_4HCO_3	0.57	-	20.5	-	52.9	50.1
NH_4Cl	-	46.5	-	31	-	27
NH_4Ac	1.02	43	21.5	47	-	49
NH_4Cl	0.73	44.8	24.0	36	42.0	21.6
NH_4Cl	0.67	-	19.7	-	-	-
NH_4Cl	0.57	-	24.0	-	-	-
NH_4Cl	0.87	-	23.1	-	-	-

experiments, however, that there is a marked rise in blood ammonia content coincident with the infusion. This clearly indicates that there is a maximum rate (or threshold) at which the liver is capable of converting ammonia to urea and beyond which toxic signs may develop.

The determination of blood ammonia is difficult procedure and is not well adaptable to clinical use, especially because of the rapid rise which occurs after shedding of blood.

SUMMARY AND DISCUSSION

The increasingly frequent use of intravenous ammonium chloride in the therapy of alkalosis and as a diuretic demands a thorough knowledge of its actions by this route of administration. In studying the pharmacology and toxicology of the intravenous injection of ammonium salts, we have observed its effects on thirty-two normal adult dogs. We have used four ammonium salts of different reaction in order to establish the toxicity on the basis of the ammonium ion rather than to investigate the alteration of acid-base equilibrium caused by the anion. We have found that the same toxicity occurred whether the CO_2 combining power was normal or in the range of acidosis. It has also become apparent that the appearance of toxicity and death is function of the rate of administration and not of the total amount of ammonium compound given. This is borne out by the following data; (a) An average rate of administration of 0.292 milliequivalents per kilograms per minute produced toxicity in eight minutes and death occurred in twelve minutes after the onset of toxicity. This is 6.48 times the rate of administration used in clinical practice by several authors. (b) An average rate of infusion

0.103 milliequivalents per kilogram per minute or 2.29 times the recommended rate, produced toxicity in 63 minutes. While death had not occurred twenty-eight minutes later when the infusion was stopped, it is likely that the death would have occurred if the intravenous injection had been allowed to continue for a longer time. (c) An average rate of 0.123 milliequivalents per kilogram per minute produced toxicity in 45 minutes and death occurred 44 minutes later. This was the lowest rate of infusion which caused death.

In the majority of cases the toxicity followed a consistent pattern characterized by irregularity of respiration, cyanosis, cardiac irregularity, initial rise and terminal fall in blood pressure, muscular twitching progressing to convulsions, hemolysis, respiratory arrest and death. However, in a smaller percentage of the cases, especially those with a high rate of infusion, the typical train of toxicity did not appear, and the animals died without previous warning signs.

Forbes and Erganian (8) reported a similar train of toxic events in one of their patients who received ammonium chloride intravenously for 18 minutes at a rate which we have calculated from their data to be 0.276 milliequivalents per kilogram per minute. They believed the toxicity was due to an extensive, rapid fall of serum pH. However, in light of our results showing identical toxic reactions whether the CO_2 combining power was rising or falling, it is evident that the probable cause was a too rapid accumulation of ammonium ions. In preliminary experiments on dogs and rabbits, the same authors found minor toxic signs with rates varying from 0.074 to 0.111 milliequivalents per kilogram per minute (our calculations from their data) continued over a 45

minute period. These data are wholly consistent with ours, indicating that the threshold for toxicity is similar for dogs, rabbits, and humans.

These studies have considerable clinical importance in light of the narrow safety margin of intravenous ammonium compounds. The fact that the rate of infusion is the determining factor in the causation of toxicity makes it of great importance to have constant supervision during the infusion. It is possible that an inadvertent speeding of the rate of infusion to two or three times the intended rate would produce toxicity if injection is allowed to continue for 45 to 60 minutes. A rate of 5-6 times the recommended dose, which is within the realm of clinical possibility might produce toxicity and death in 15-20 minutes. Furthermore, there is no valid physiologic reason to use a high rate of administration since the object sought in the correction of edema is to have the displaced sodium ions pass through the glomerulus and be excreted in the urine. No matter what the rate of infusion, each chloride ion released from ammonium chloride, when excreted, will take with it a sodium ion. In alkalosis, the addition of each chloride ion will cause a shift of the buffer system toward the normal state no matter how rapidly they are infused. Thus in the treatment of edema or alkalosis with ammonium chloride there is no advantage to run the risk of toxicity by attempting to lower the blood CO_2 combining power too rapidly.

Forbes and Organian (8)consider it doubtful that an increase in ammonia content of the systemic circulation has ever been detected, even after ingestion of large quantities of ammonium salts. However, Kaprowski and Uninski (13) have shown in dogs, that blood ammonia and urea rises markedly after oral administration of ammonium chloride and we have shown that there is a more marked rise after intravenous administration of

ammonium chloride. Fiske and Karsner (7) state that conversion of ammonia to urea in the liver does not seem to be hampered by hepatic damage produced by chloroform and phosphorus. Their method of determining blood ammonia was cumbersome and probably not accurate, their normal level being much higher than that found by Conway (2) by more modern methods. This author has also shown that the normal circulating human blood ammonia is virtually zero, and that the blood ammonia previously reported was formed after shedding. In view of this fact, an increase of blood ammonia of a few micrograms per milliliter assumes new importance, and the question of ammonia metabolism in the presence of hepatic damage should be re-examined. Until such a study has been performed with modern methods, it would seem wise to regard severe liver damage as a contraindication to the intravenous use of ammonium chloride.

We have shown in our experiments by using four ammonium salts of different reaction that the toxicity described is due to the ammonium ion and not to changes in CO_2 combining power. For instance, the rates of infusion of ammonium bicarbonate which causes practically no change in CO_2 combining power, and of ammonium chloride which causes a marked fall, correspond closely for the same effect.

CONCLUSIONS

1. Physiologically Acid, neutral, or basic ammonium salts by vein are equally capable of causing toxicity and death.
2. A rate of administration 2.3 times that which has been used clinically produced toxicity in 65 minutes and a rate 6.43 times the recommended rate produced toxicity in 8 minutes.
3. Blood ammonia content rises markedly during rapid infusion of ammonium salts.
4. Ammonium chloride by vein causes a lowering of the CO_2 power.

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