Blood Metabolites in the Hyperammonemic Pig (34742)

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Biochemical changes in the blood, liver, and brain due to ammonia toxicity have been described in some detail for the rat (1–4), although little such information is available for the pig and other simple-stomach animals. Associated with ammonia toxicity are elevated levels of blood and liver glucose, lactate, pyruvate, glutamate, oxaloacetate, and a buildup in the brain of lactate and pyruvate. With these are concurrent reductions in reduced pyridine nucleotides and alpha-ketoglutarate in the liver.

The purpose of the present studies was to determine the chronological changes of some of the blood metabolites mentioned above, together with blood pentoses, ketone bodies, and plasma calcium, phosphorus, and magnesium in the pig under induced hyperammonemia, and the effect, if any, of therapeutic doses of magnesium salt on the response to injected ammonia.

Materials and Methods. Expt. 1. Eight Yorkshire suckling gilts weighing 3.5 to 4.0 kg were divided into groups of four animals each.

Five ml of blood were withdrawn from the anterior vena cava of each pig in the two groups and collected in heparinized centrifuge tubes. These served as zero-time blood samples.

Ammonium carbonate (150 mg/kg of body wt administered in 20% aqueous solution) was injected intraperitoneally into each pig in the two groups. In the group that received magnesium, 25 mg of magnesium sulfate (5% aqueous solution)/kg of body weight were injected intraperitoneally immediately after the ammonium salt was administered.

Blood samples of 5 ml each were then taken from each pig at 5, 15, 30, 60, and 120 min after ammonium carbonate injection. Protein-free filtrates of all blood samples were prepared immediately after sampling by the method of Folin and Wu (5). Total blood ammonia, glucose, lactate, pentoses, and ketone bodies were determined (6–10). Plasma from the original blood samples was analyzed for magnesium, phosphorus, and calcium by atomic absorption spectrophotometry with lanthanum chloride added to eliminate the interference of phosphorus in the Mg and Ca determinations.

Expt. 2. Seven Yorkshire suckling pigs weighing 5.0–7.0 kg were divided into two groups of two controls and 5 ammonia-injected animals. Blood (8 ml) was obtained as previously and at 10, 30, 60, and 120 min after ammonium carbonate administration or physiologocal saline administration. The same blood analyses were made as previously except that no plasma mineral analyses were made and blood pyruvate (13) and dehydroxyacetone-*P* (14) were determined.

Results. Expt. 1. The physical manifestations of the ammonia toxic, well-nourished pig, heretofore undescribed in the literature, remarkably resembled those described for grass staggers of cattle by Sjollema (11). These included nervousness, restlessness, and unsteady gait, muscle twitching and gnashing of teeth followed by vomiting. Within 10 min of intraperitoneal administration of ammonium carbonate, the animal entered into convulsive tetany which seemed to be precipitated by the animal's extreme hypersensitivity, especially to noise. (An ammonia toxic mouse was so excitable that it could be made to jump repeatedly a few inches off the floor with each snap of the fingers of the experimenter despite the otherwise limp condition of the animal.) The limbs of the pig were stretched out and the head flung back. The

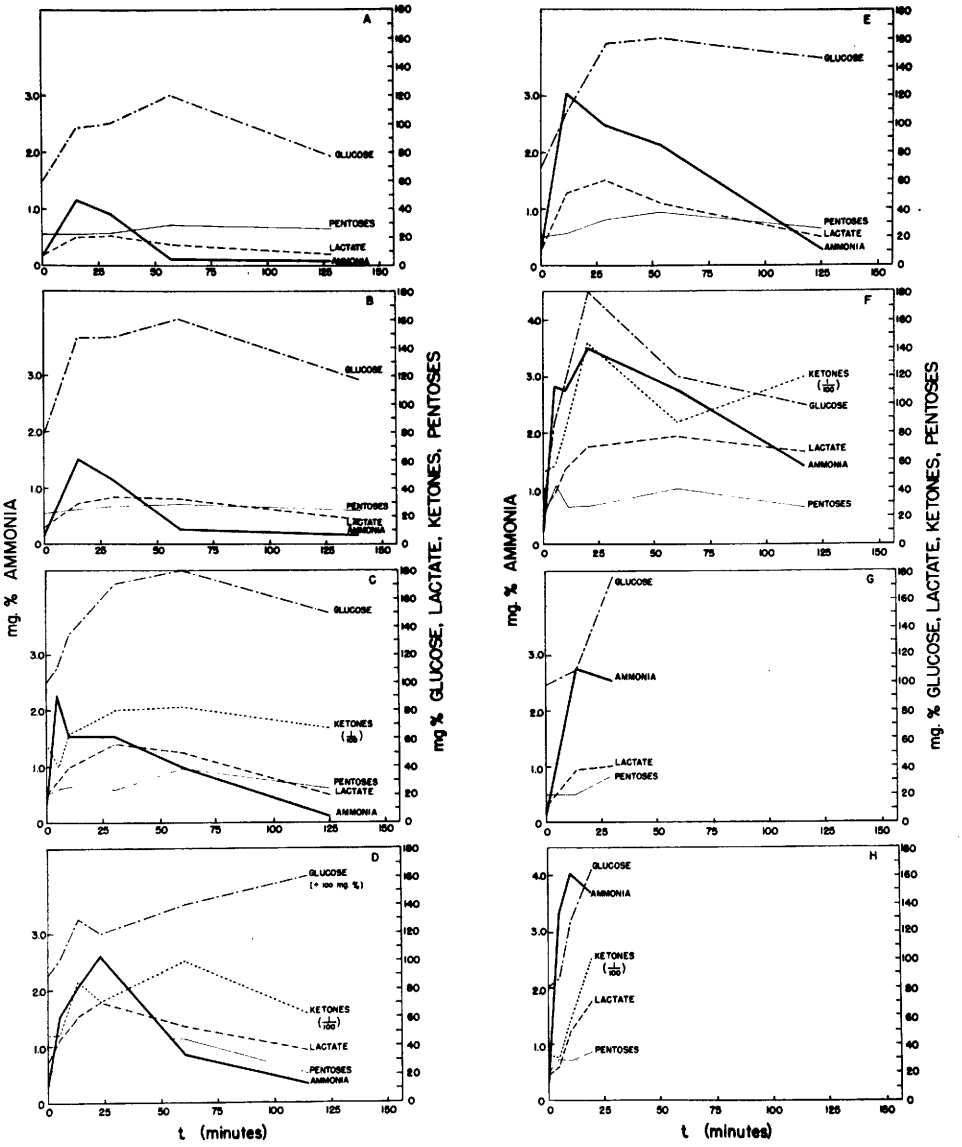

FIG. 1. Chronological changes of pig blood glucose, lactate, pentoses, and ketone bodies with changes in peripheral blood ammonia. Pigs D, E, G, and H received intraperitoneal doses of magnesium sulfate.

eyes were anxious and wild-looking. The animal repeatedly entered this extreme state, which lasted no more than 5 sec each time, and then into coma. Death resulted in 2 of the 8 pigs. Recovery was complete after 2 hr in survivors.

A rise in peripheral blood ammonia was associated with a rise in blood glucose and lactate, confirming earlier work (1–4) (Fig. 1). Blood pentoses and total ketone bodies were also elevated. Hyperammonemia was further associated with an apparent rise in plasma magnesium and phosphorus. Plasma calcium, however, was not affected (Fig. 2). Results of this study are summarized in Table I. Magnesium sulfate at the level administered did not have any effect on any of the levels of organic metabolites in the blood.

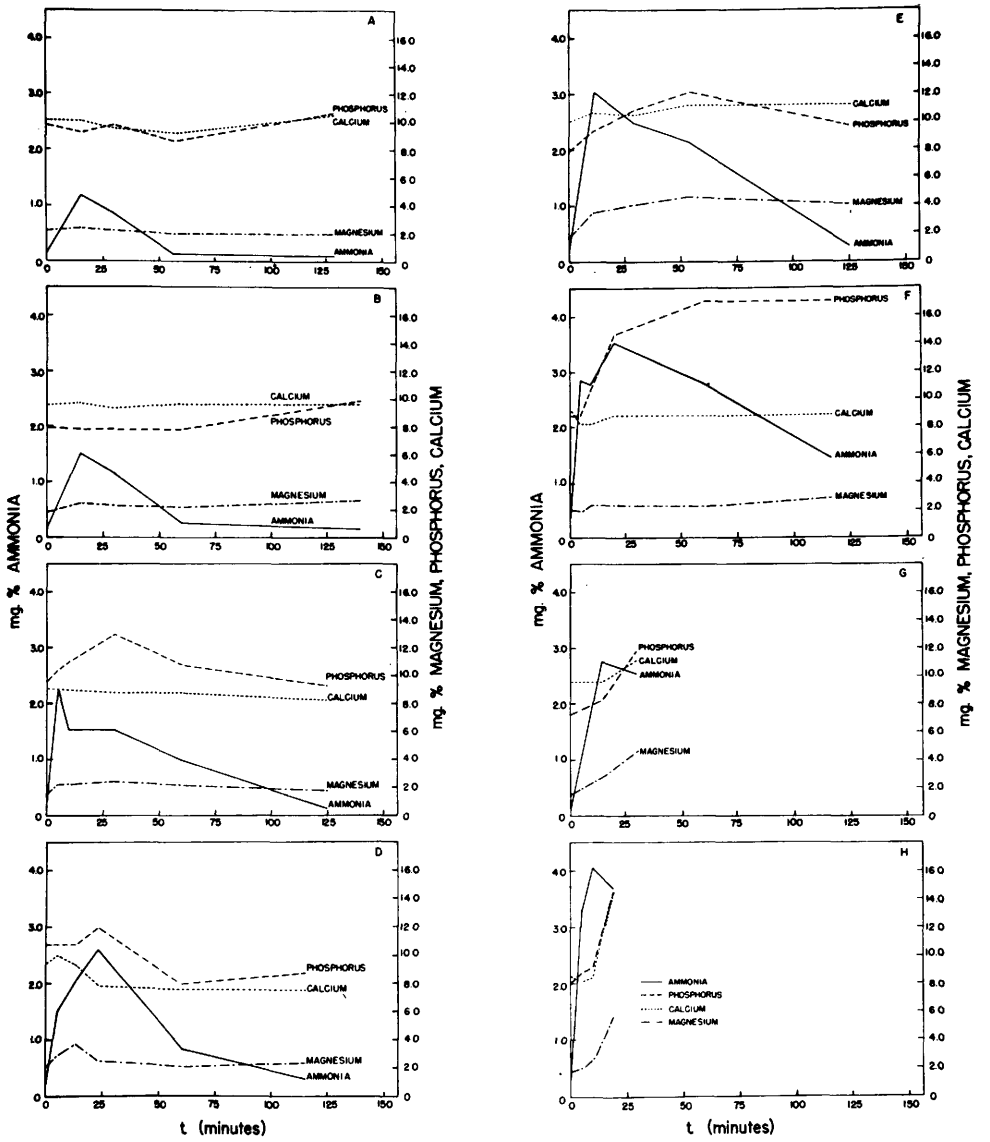

FIG. 2. Chronological changes of pig plasma calcium, phosphorus, and magnesium with changes in peripheral blood ammonia. Pigs D, E, G, and H received intraperitoneal doses of magnesium sulfate.

Expt. 2. The physical manifestations of ammonia toxicity were the same as those observed in Expt. 1. The changes in blood metabolites following ammonia administration are shown in Table II. The rise in all components measured was similar to that of Expt. 1. Blood pyruvate and dihydroxyacetone-*P* also rose sharply following ammonia administration. Pyruvate reached a peak of 160 to 200% above the initial level and dihydroxyacetone-*P* reached a peak of 200 to 300% above the initial level after 30 to 60 min in survivors. Two pigs died approximately 30 min after ammonia administration and one died after 120 min.

Discussion. The changes in blood organic metabolites associated with hyperammonemia suggest the breakdown of normal metabolic mechanisms in the animal body. The buildup of glucose and lactate suggests an inhibition

TABLE I. Blood and Plasma Metabolites of Pigs with Induced Hyperammonia (Exp. 1).

Metabolite	Before ammonia (mg/100 ml)	Time after ammonia (min)[a]; (mg/100 ml)				
		5	15	30	60	120
Blood						
Ammonia[b]	0.15	2.48	2.32	2.29	1.19	0.40
Lactate[b]	16.5	31.2	41.3	51.6	45.5	28.2
Glucose[b]	100	145	146	177	180	158
Pentoses[b]	20.8	30.3	25.1	31.0	36.7	24.7
Ketones[b]	0.47	0.44	0.72	0.99	0.91	0.84
Plasma						
Mg[b]	1.88	2.26	2.77	3.31	2.57	2.65
P[c]	8.75	9.70	9.55	12.86	10.68	10.88
Ca	9.44	8.88	9.36	10.05	9.17	9.28

[a] 0–30 min represent means from 8 animals, 60 and 120 min represent means from 6 animals.
[b] Significant rise after ammonia administration ($p < 0.005$); [c] ($p < 0.10$).

of the TCA cycle. This could be due to an inhibition of the decarboxylation of pyruvate through the uncoupling effect of NH_3 on oxidative phosphorylation demonstrated *in vitro* (1). The concurrent rise in ketone bodies lends support to this suggestion, since the hyperammonemic animal would then depend heavily on fat catabolism to meet its energy needs. Further, the buildup of blood pentoses may indicate an increased metabolism of glucose via the pentose phosphate pathway. This effect due to ammonia has been reported for *in vitro* systems earlier (12).

TABLE II. Blood Metabolites of Pigs with Induced Hyperammonemia (Exp. 2).

Metabolite	Before ammonia (mg/100 ml)	Time after ammonia (min); (mg/100 ml)			
		10	30	60	120
		Control pigs[a]			
Ammonia	0.14	0.16	0.13	0.13	0.14
Lactate	21	18	17	16	13
Pyruvate	1.19	1.38	1.38	1.23	1.05
Glucose	82	83	79	78	81
Pentoses	25.6	25.7	24.2	23.9	24.0
Ketones	0.45	0.46	0.45	0.45	0.46
Dihydroxyacetone-*P*	4.26	4.29	4.29	4.45	4.68
		Ammonia intoxicated pigs[b]			
Ammonia	0.14	2.22	3.78	2.11	2.22
Lactate	26	34	42	53	63
Pyruvate	1.39	1.74	2.10	2.46	2.42
Glucose	90	138	195	193	171
Pentoses	24.5	30.7	35.3	34.4	34.9
Ketones	0.43	0.52	0.74	0.71	0.89
Dehydroxyacetone-*P*	3.93	5.49	7.49	7.85	8.84

[a] Each mean represents 2 animals given saline (no ammonia).
[b] 0, 10, and 30 min represents 5 animals; 60 and 120 min represents 3 animals.

Summary. The chronological events occurring in blood after intoxicating doses of ammonium carbonate were studied using the suckling pig. Physical manifestations included vomiting, hyperventilation, hypersensitivity to sound, convulsions, and tetany followed by death in some cases. Blood glucose, dihydroxyacetone-*P*, pyruvate and lactate were raised indicating failure to decarboxylate pyruvate. Elevated blood ketone bodies indicated increased fat catabolism as a result. Consistent rise in blood pentose may either be due to an inhibition of the pentose phosphate pathway or an increased metabolism of glucose via this pathway to make up for reducing power lost through impairment of the TCA cycle.

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