stimulated growth when fed at a level equivalent to 74.25 mg of 3-nitro/kg diet, with the dimer somewhat more effective. There was little difference between the 2 when fed at a level equivalent to 198 mg 3-nitro/kg diet. Liver arsenic stores were greater in chicks fed the dimer at each of the 3 levels of comparison. Administered as a single oral dose at levels approximating 100 and 200 mg/kg body weight, survival out of 10 chicks treated was 7 and 1 respectively, for 3-nitro and 8 and 0 for the dimer.

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Hypoglycemic Activity of Tris Buffer in Man and Dog. (25194)

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Tris(hydroxymethyl)aminomethane [$(CH_2OH)_3 \cdot C\text{-}NH_2$; tris buffer, THAM] has been used widely in biochemical work as a buffer. Nahas(1) discovered that it can be safely infused into dogs and corrects profound respiratory acidosis. Stimulated by this observation, our laboratory participated in a study of the alkalinizing effects of intravenous tris in healthy, fasting adult man. Since little was known of *in vivo* properties of tris, we set out to characterize its effects in terms of chemical measurements available in our laboratory. We were surprised to find a hypoglycemic effect.

Studies in man. After an oral water load, 4 subjects breathed room air for about half an hour and then 2.3 or 3.4% CO_2 for a similar period. Intravenous tris (0.3M in 0.03M NaCl and 0.005M KCl) was given immediately thereafter, during CO_2 breathing, over 30 to 64 minutes. A recovery period followed. Arterial blood was sampled from an indwelling femoral cannula. Serum glucose(2) fell distinctly (Table I) in 3 subjects during and at the end of tris infusion. Hypoglycemia occurred in J.F., who received the largest dose (8.8 mM/kg or 0.49 ml/kg/min over 60 minutes). This was accompanied by hunger and profuse sweating superseded by other toxic manifestations which persisted for 24 hours. Glucose returned toward pre-infusion values within 57 minutes after infusion in J.F. Glucose fell substantially in S.B., 2.8 mM tris/kg (0.19 ml/kg/min over 49 minutes) and in J.D., 4.1 mM/kg (0.21 ml/kg/min over 64 minutes). The fall in J.L. to 65 mg/100 ml occurred 26 minutes after tris infusion was over (3.0 mM/kg or 0.33 ml/kg/min for 30 minutes). Serum inorganic phosphate fell progressively with glucose in J.F. from 3.6 to 1.9 mg/100 ml, from 3.5 to 2.9 mg/100 ml in J.D., from 3.8 to 3.3 mg/100 ml in J.L., but was unchanged in S.B. Serum potassium did not fall in any of the subjects. Other chemical and physiologic observations are reported elsewhere(3,4).

Studies in dogs. Tris was then given intravenously to 3 fasting, anesthetized (pentobarbital), heparinized, female dogs during room air breathing. 0.3M saline was infused at 0.5 ml/kg/minute over 1 hour; then 0.3M tris (in 0.03M NaCl and 0.005M KCl) at the same rate (slow infusion, Table II) for 2½ to 3 hours followed by 1 hour at 0.7 to 1.0 ml/kg/minute (fast infusion, Table II). Femoral arterial serum glucose concentrations rose during anesthesia and manipulation of 2 of the animals and then tended to remain unchanged or return to pre-anesthetic levels during saline infusion. Glucose declined during slow infusion of tris in all dogs. Values fell to below 50 mg/100 ml in 2 dogs. Rapid infusion produced a precipitous further reduc-

tion in serum glucose in all dogs. Serum inorganic phosphate fell to less than 50% of control values in all dogs, during tris injection, reaching levels consistently below 1.0 mg/100 ml in 2 of the animals. This fall probably signified a net transfer of phosphate from extracellular fluid to cells, since urinary phosphate excretion decreased. There was no definite trend in serum potassium.

Other findings related to tris administration in the dogs include: Blood pH rose by 0.24 to 0.30 units and plasma CO_2 content increased by 4 to 9 mM/L. Serum sodium fell by 14 to 27 and chloride by 17 mM/L. Arterial blood pressure rose, and remained high, after the first 2 to 2½ hours of tris. Urine flow and excretion of sodium, chloride, and potassium rose.

Glycosuria did not develop during tris injection into either man or dog. *In vitro* additions of tris greatly exceeding estimated concentrations *in vivo* did not artifactually lower glucose or phosphate.

Discussion. The present experiments do not establish whether the fall in serum glucose induced by tris is mediated by insulin, is a specific hypoglycemic effect independent of insulin, or represents diminished gluconeogenesis. Work is in progress to examine these possibilities. The phosphate fall resembles that following glucose and/or insulin administration. Lack of fall in serum potassium may reflect the small load of potassium given with tris rather than a difference from the potassium-lowering tendency of glucose and insulin.

TABLE I. Arterial Serum Glucose in 4 Adult Males. (No. in parentheses represent min. from start of tris or recovery period.)

	Glucose, mg/100 ml		
S.B. 88.5 kg	J.D. 83.2 kg	J.F. 71.7 kg	J.L. 75.5 kg
	Room air		
91	102	100	75
	CO_2		
97	111	89	80
	Tris and CO_2		
78 (30′)	90 (32′)	55 (39′)	77 (18′)
74 (49′)	80 (64′)	52 (56′)	76 (25′)
	Recovery		
75 (60′)	86 (98′)	79 (57′)	72 (5′)
		89(122′)	65 (26′)

TABLE II. Arterial Serum Glucose in 3 Dogs.

	Glucose, mg/100 ml		
Time, min.	#110 14.7 kg	#111 22.2 kg	#112 13.2 kg
	Pre-anesthesia		
		92	114
	Pre-NaCl (2-3 hr later)		
	100	131	122
	NaCl infusion		
30	94		114
60	93	128	106
	Tris infusion (slow)		
30	84	110	103
60	73	94	84
90	77	85	88
120	53	69	78
140	45*	53	79
160		37	79
180		30	78
	Tris infusion (fast)		
15		26	60
30	13	25	60
45		10	43

* Slow infusion ends.

The hypoglycemic action of tris in the intact organism has broad implications. Clinically, it emphasizes one of the possible sources of toxic side effects if the amine is used for treating acidosis. Nevertheless, it is conceivable that this or related compounds may prove useful in treating diabetes mellitus or diabetic acidosis. Moreover, studies of the relation of tris and similar compounds to carbohydrate metabolism both *in vivo* and *in vitro* may provide a fresh approach to problems of carbohydrate regulation.

Summary. Tris(hydroxymethyl)aminomethane produced significant hypoglycemia in man and dog. The nature of this phenomenon remains to be determined.

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