

Urinary Output of Nitrogen as Influenced by Intravenous Injection of a Mixture of Amino-Acids.

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Evidence that a mixture of amino-acids injected intravenously produced rapid regeneration of serum protein was recorded previously.¹ The present communication deals with observations on the urinary excretion of nitrogen as influenced by the administration of amino-acids in this way. Previous experiments by Henriques and Andersen² have indicated that a positive nitrogen balance may be attained even when all of the nitrogen intake was injected intravenously in the form of an enzymatic digest of meat. These authors used one goat, but the preparation was not completely hydrolyzed to amino-acids inasmuch as it contained 15% of its nitrogen in the form of peptides.

In the present experiments the mixture of amino-acids was obtained by the acid hydrolysis of casein, as previously described.¹ Such a preparation contains no tryptophane, which is destroyed during the hydrolysis and is low in cystine. This incomplete basic mixture, however, served as a control for the observations in which 2% of each of these 2 essential amino-acids were added to form the "complete" mixture.†

Healthy, adult, short-haired dogs were used. They were kept in metabolism cages, allowed water *ad libidum*, starved for a few days and then given, twice daily, by gavage a solution of 12½% sucrose in normal saline. The amount of sucrose administered in this way was calculated to yield about 50 to 60 calories per kilo of body weight. The same amount was given daily in each case. This procedure tended to spare protein breakdown and thus produced a much lower nitrogen output in the urine than occurs during starvation. Moreover, it acted as such a good diuretic that large, relatively

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¹ Elman, R., *PROC. SOC. EXP. BIOL. AND MED.*, 1937, **36**, 867.

² Henriques, V., and Andersen, A. C., *Z. f. physiol. chemie*, 1913, **88**, 357.

† The hydrolyzed casein was kindly furnished by Mead, Johnson and Company. The added tryptophane and cystine were obtained from Eastman Kodak Company. Each of the added amino-acids was levorotatory.

uniform amounts of urine were passed each 24 hours; catheterization was thus deemed unnecessary. The feces under the conditions of the experiment were scanty and infrequent; their nitrogen content was assumed to be constant and slight and they were therefore not saved for analysis. The 24-hour urine was collected in receptacles containing toluol to prevent fermentation and sufficient acid to neutralize any ammonia. All experiments were carried out in consecutive 3-day periods. The total nitrogen of the urine was determined by direct digestion and Nesslerization. Following the preliminary starvation period, the dogs were observed for from 3 to 7 days during which period the output of urinary nitrogen dropped considerably; experiments were begun only after the urinary nitrogen approached a relatively uniform level. The amino-acids were given in a 10% solution containing also 10% glucose. Ten to 20 cc. were injected intravenously each hour until the total daily dose was given; this usually required 6 to 10 hours and produced, in no case, any evidence of toxicity. Previous observations had shown that with this rate of injection very little of the amino-acids appeared in the urine (*i. e.*, under 10%).

The findings, as shown in Table I, represent 15 days' observation on 2 dogs; they were chosen from experiments on 7 dogs, all of which showed similar results. It will be noted that N-balance was immediately achieved during the periods in which the complete mixture of amino-acids was injected; this was possible even though the amount of nitrogen administered was slightly in excess of the amount excreted in the preceding period. Thus, in spite of the large amount of nitrogen injected there resulted only a slight increase in the urinary nitrogen excreted, indicating a very high degree of retention of the injected amino-acids. In dog B70 3 gm. of nitrogen were excreted each day during the first 3 days; 3.11 gm. were excreted daily the next 3 days during which 3.16 gm. of nitrogen as amino-acids were injected. In dog B9 the results were similar, *i. e.*, while 1.58 gm. of nitrogen were being given daily the output was the same, whereas in the 3 preceding days when no amino-acids were given the output was only slightly lower, 1.47 gm. The immediate achievement of nitrogen balance has not been the case in many experiments by others in which protein or amino-acids were fed by mouth. The authors mentioned above,² however, did achieve such an immediate effect; they are apparently the only other observers who studied nitrogen balance during the intravenous administration of amino-acids.

In contrast to these findings with the complete mixture of amino-

TABLE I.

Day	Urinary Output			N-Intake Daily Injections Amino acids (gm. N)	N-Balance	Remarks
	Amt. (cc.)	Total N (gm.)	Daily Aver. (gm. N)			
1	900	2.79				Dog B70
2	600	2.52				Wt. 10.1 kg.
3	820	3.70	3.00	0.0	-3.00	125 gm. sucrose daily
4	850	3.21				Amino-acid mixture
5	1200	3.71				containing 2% tryptophane and added
6	1100	2.40	3.11	3.16	+ .05	cystine
7	1000	2.50				
8	850	2.47				
9	810	2.51	2.50	0.0	-2.50	
10	750	3.45				Incomplete (basic)
11	850	4.25				amino-acid mixture
12	900	3.33	3.68	3.16	- .52	
13	830	3.32				
14	750	1.80				
15	670	2.14	2.42	0.0	-2.42	Wt. 8.6 kg.
1	600	1.44				Dog B9
2	600	1.14				Wt. 6.6 kg.
3	800	1.52	1.37	0.0	-1.37	100 gm. sucrose daily
4	1100	2.31				Incomplete (basic)
5	1000	1.90				amino-acid mixture
6	1000	2.00	2.07	1.29	- .78	
7	950	1.90				
8	490	1.12				
9	700	1.40	1.47	0.0	-1.47	
10	1000	1.60				Amino-acid mixture
11	1200	1.56				containing 2% tryptophane and added
12	1000	1.60	1.58	1.58	0.0	cystine
13	700	1.40				
14	900	1.89				
15	750	1.72	1.67	0.0	-1.67	Wt. 6.0 kg.

acids are those with the basic mixture in which no tryptophane or cystine was added. In neither dog was N-balance achieved though the amount of nitrogen injected was about the same. Indeed, of the injected amino-acids only about one-half was retained in one case and but four-fifths in the other. It is of interest to note too that the increased nitrogen output which occurred during the days of injection persisted for the next 24 hours into the period during which no injections were given. This did not occur when the complete mixture was injected.

It should be emphasized that these observations are not concerned with the biological value of hydrolyzed casein inasmuch as a condition of minimum endogenous nitrogen metabolism was probably not achieved in these dogs. Moreover, the period of observation was too short for such a study. The experiments are concerned only with the question as to whether amino acids injected intravenously this way are efficiently utilized by the body, particularly in the synthesis of tissue protein. It seems clear that if the amino-acids were retained as such that this would take place at approximately the same degree regardless of whether or not the mixture contained the 2% added tryptophane and cystine. That a high degree of retention occurred only with the complete mixture is evidence that synthesis of protein had probably taken place.

Summary. Adult dogs, fed only with sucrose, were immediately brought into N-equilibrium upon intravenous injection of a complete mixture of amino-acids combined with glucose. This appears to demonstrate a high degree of nitrogen retention particularly since nitrogen balance was attained with the administration of an amount of nitrogen but slightly in excess of the previous level of excretion. This was impossible if the mixture did not contain added tryptophane and cystine; under such conditions a large part of the injected nitrogen appeared in the urine. These observations would seem to indicate that the mixture of amino-acids used, as prepared by the acid hydrolysis of casein plus 2% tryptophane and added cystine, is efficiently utilized by the body, probably in the active synthesis of body protein.

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The Blood Diastase in Acutely Perforating Peptic Ulcers.*

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Meyer and Antman¹ recently reviewed the meagre literature on the blood and urinary diastase in peptic ulcers. The results were inconclusive. They reported 3 cases of high urinary diastase in patients with peptic ulcers involving the pancreas. One case was

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¹ Meyer, K. A., and Antman, L., *Am. J. Surg.*, 1936, **33**, 307.