

The excretion of urea in relation to protein intake and diuresis

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SUMMARY

The urea excretion of 15 children on a low protein diet and 15 orphans as normal controls was studied at different levels of urine flow.

In the groups of children on a low protein diet the urea excretion remained approximately constant with increasing urine flow, while in the normal controls a significant increase of urea excretion was observed with rising diuresis. It is concluded that comparative studies between groups on different levels of protein intake are best undertaken at high urine flows to obtain maximum differences in urea excretion.

INTRODUCTION

For many years it has been known that the daily urinary nitrogen excretion varies with the nitrogen intake (1). Therefore urinary total nitrogen or urea-nitrogen excretion has been widely used as an indicator of protein intake. While we would expect a 24 hour urinary excretion to reflect protein intake, the determination of nitrogen in 3 or 6 hour samples would add the problem of diurnal variation. Experiments have shown that a person placed on a very low protein diet loses nitrogen according to the previous level of protein intake, after which nitrogen excretion decreases mainly due to a reduction in urea excretion (2). Therefore the determination of fasting urea excretion avoids the problem of diurnal variations due to previous protein intake, and has proven to be valuable in nutri-

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tion surveys. Only relatively recently nutritionists drew attention to the fact long known among nephrologists that urea excretion varies considerably with diuresis. Schmidt-Nielsen (3) observed in sheep on a low protein intake a marked increase of the urea clearance with increasing urine flows. In sheep on a normal protein intake the urea excretion was constant over a wide range of urine flows. The same phenomenon was observed by Murdaugh (4) who found in man after 5 weeks of low protein intake a marked decrease in the fraction of filtered urea excreted at low urine flows, which was however not apparent during maximal water diuresis. Following these observations Arroyave (5, 6) studied the urea excretion of children with a high and low protein intake in relation to urine flow rates with the same results and concluded that when the basal urea excretion is measured to distinguish between population groups of different protein intakes, the water intake should be restricted to produce maximum differences in urea output.

The findings of the above authors are not fully supported by studies of other workers. Rehberg (7) found in normal man that the fraction of filtered urea which is excreted decreases in a regular manner with decreasing urine flow. Shannon (8) confirmed these findings in dogs. Kenney (9) found in contrast to Arroyave and Schmidt-Nielsen (10) a maximum difference in urea excretion between West-Africans and Europeans at high urine flows while with a low urine production the difference became non-significant. The considerable controversy regarding the excretion of urea in relation to protein intake and diuresis led us to study the problem in two extreme nutritional groups of children.

METHODS AND MATERIALS

Subjects

The sample included 15 children of squatters in Kenya on a low protein intake and 15 children in a Nairobi orphanage on a typical European diet as normal controls. The children in both groups were of approximately the same age averaging between 4 and 5 years. The generally low protein intake in the group of squatters is supported by some qualitative data obtained from the general population. When 82 mothers were asked what they fed their children up to 6 years on the pre-

vious day 68 answered maize, 64 potatoes, 39 bananas, 24 legumes, 4 milk and 2 vegetables, 2 tinned food and 1 each eggs or pumkin (11). Similarly the quality of the protein eaten was significantly lower in the squatter children, as measured by the urinary inorganic sulfate sulfur/creatinine ratio. Taking the data of the 3 test days together a mean value of 428 ± 166 $\mu\text{g}/\text{mg}$ was found in the squatters as compared to 690 ± 331 $\mu\text{g}/\text{mg}$ in the orphans. The difference between the two groups is significant at the 0.1% level.

In agreement with these findings is the observation that out of the 15 squatter children 9 had a marked depigmentation of the hair as a sign of PCM. Though weight for age is indicative of past malnutrition and not related to recent food intake a comparison of the two groups is strongly suggestive that protein intake had been low among the squatter children. While the mean weight for age of the squatters was $83.3\% \pm 8.3$; the orphans averaged $94.9\% \pm 9.2$ ($P < 0.1\%$). It can therefore be concluded that the past nutritional status and the protein ingestion at time of examination were significantly lower in the squatters than in the orphans.

Methods

Urine was collected each day on three successive days. At the beginning of each collection period at 7 a. m. all children emptied their bladder completely and again after three hours. No food was allowed prior to and during the collection of the timed urine samples.

On the first day of the study the children of both groups were given no water. On the second day the children received a water-load of 10 ml per kg body weight at the beginning of the collection period. On the third day the same procedure was repeated with a water-load of 20 ml per kg body weight. Thus different levels of water diuresis were obtained.

Chemical Methods

Creatinine was estimated in an autoanalyzer using the method of Folin and Wu (12). The urea was also determined in the autoanalyzer applying a method described by Wootton (13). The osmolarity of each sample was determined by boiling point elevation (14). Urinary inorganic sulfate sulfur was estimated using a method first published by Berglund and Sörbo (15).

RESULTS

The results of the water-load are shown in the Table I. The urine flows observed in our study resemble those observed by Arroyave (5, 6) under similar conditions. Remarkable is the high urine flow in the group on a low protein intake (squatters) accompanied by a low urinary osmolarity, when compared with the normal controls (orphanage). The creatinine excretion was almost constant with the increasing water-load. The increases from 20.2 mg/3 hr to 23.8 mg/3 hr in the groups of squatters and from 24.1 mg/3 hr to 33.2 mg/3 hr with increasing diuresis were not statistically significant. With regard to the excretion of urea in relation to protein intake and water diuresis the results are in contrast with the reported findings of Arroyave (5, 6), Murdaugh (4) and Schmidt-Nielsen (10). The absolute urea-N excretion over 3 hours remained constant with increasing diuresis within small limits in the group on a low protein diet. In the normal controls however the level of urea increased from a low of 314 mg/3 hr at low urine flow to as high as 557 mg/3 hr in extreme water diuresis and was significant at the 5% level. To avoid a possible error through incomplete collection of the 3 hour urine samples the urea-N excretion was also expressed in grams per grams of creatinine in the same samples, making use of the relative constancy of urinary creatinine excretion. The results are strikingly similar to those mentioned above for the absolute urea excretion. Again the urea-N creatinine ratio was found constant within a very small range in the group of squatters while in the group on a high protein intake with increasing diuresis a rising ratio from 12.9 to 16.6 was observed at a significant level of 5%.

DISCUSSION

The fact that without a water-load the urine flow was higher in the group of children on a low protein diet may have been caused by two factors. The squatters lived in a cold climate on the slopes of Mount Kenya while the orphans lived in Nairobi at moderate temperatures which increases the extrarenal water loss. On the other hand it is a well established fact that urea plays a very important part in urine concentration. Levinsky (17) found that the passive accumulation of

TABLE 1

URINARY EXCRETION PATTERN OF UREA IN RELATION TO DIURESIS

		Water-load in ml per kg body weight						Significance (16) of differences	
		0		10		20		between the 3 test days	
		\bar{X}	SD	\bar{X}	SD	\bar{X}	SD	F	P
Osmolarity (mosmol/l)	Squatters	234	124	138	71	76	21		
	Orphanage	392	159	163	84	102	44		
Urine flow (ml/min)	Squatters	0.63	0.26	1.13	0.39	1.89	0.66		
	Orphanage	0.29	0.17	0.86	0.39	1.46	0.37		
Creatinine (mg/3 hr)	Squatters	20.2	6.3	22.5	9.9	23.8	7.4	2.245	ns
	Orphanage	24.1	8.9	30.3	12.1	33.2	12.6	1.995	ns
Urea-N (mg/3 hr)	Squatters	215	96	212	96	191	54	0.601	ns
	Orphanage	314	150	462	184	557	342	5.042	0.05
Urea-N Creatinine	Squatters	10.6	4.6	10.0	3.4	9.0	2.8	1.213	ns
	Orphanage	12.9	3.2	15.7	3.6	16.6	6.2	4.426	0.05

urea in the medullary interstitial fluid increases the maximum urine concentrations which the kidney can obtain. This would also explain the lower concentrations and higher urine flows under thirsting conditions in the groups of squatters, as with a low protein intake the renal urea concentration is also reduced.

As expected the urea-N excretion per 3 hr in squatters was well below the values found in orphans. If averages are calculated from the three test days a mean value of 9.9 ± 3.7 mg/mg is found for the squatters and of 15.1 ± 4.7 mg/mg for the orphans. The difference is significant at the 0.1%. The urea excretion pattern observed however was not in agreement with the findings of Arroyave (5, 6), Schmidt-Nielsen (10) and Murdaugh (4). While these authors found a constant urea excretion with increasing diuresis in normal persons and a rising excretion of urea with rising diuresis in individuals on a low protein diet we found the opposite. Schmidt-Nielsen (10) and Arroyave (5) therefore both state the difference of urea excretion on high and low protein intake is more pronounced when the urea clearance is measured during low urine flow. Our study like that of Kenney (9) rather suggest a maximum difference during extreme diuresis.

Our observation that in children on a high protein diet the urea excretion increased under diuresis is in full agreement with the findings of Rehberg (7) and Shannon (8) who established the theory that the mechanism for renal excretion of urea in mammals can basically be regarded as consisting of glomerular filtration and passive back diffusion in the tubules. On the other hand however our results in children on a low protein diet can not be explained by this theory. Schmidt-Nielsen (18) first drew attention to the possibility of tubular regulation of urea excretion and now available evidence leads almost inescapably to the conclusion that active transport of urea out of medullary collecting ducts can occur under certain conditions in the mammalian kidney, which is apparently most readily demonstrated in protein depleted animals. Lassiter (19) showed that in diuretic protein depleted rats urea is reabsorbed from collecting ducts against its concentration gradient. Qualitively similar evidence was provided by Clapp (20) in rats undergoing a more intense diuresis and Ulrich (21) more specifically states that in normal rats the urea

transport across the collecting duct wall is passive while in rats on a low protein diet an active outward transport is superimpose. These studies could provide an explanation for the almost constant level of urea excretion with increasing diuresis in the groups of squatters on a low protein diet.

Our investigation cannot provide an answer to the controversy on the mechanism of urea excretion in the mammalian kidney but it draws attention to the fact that urea excretion may vary considerably with diuresis. This study suggests that comparative studies between groups of different levels of protein intake are best undertaken at high urine flows to obtain maximum differences in urea excretion. The striking contrast of our findings to those of Murdaugh (4) may find an explanation in the differences of experimental conditions. Murdaugh conducted his study in adults in comparatively acute protein depletion while our subjects were children suffering from very prolonged protein deficiency. With the data available no satisfactory explanation can be given for the difference of our findings with those of Arroyave (5). Both field studies in which the results were different from those of Arroyave (5) were conducted in Africa. Kenney (9) compared a group of Africans with a group of Europeans. While we compared two groups of Africans one of which however on a European diet. Racial difference are therefore unlikely to provide an explanation for the differing excretion patterns.

Also climatic conditions are more different between East and West Africa than between East Africa and Guatemala. One possibility might be that the diet of the groups studied in Africa (European vs. African diet) was qualitatively more different than the diet of the groups studied in Guatemala (high socio-economic vs. low socio-economic group diet).

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