EXCRETION OF SULPHUR IN URINE OF INDIANS IN GUJARAT

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"Measurement of a day's output in urine, of total sulphur gives an approximate indication of the quantity of protein that the subject is injesting" (Harrison, 1939). The excretion of sulphur in urine of Indians has been examined by very few workers. Ray and Ganguli (1938) reported values of urinary sulphur excretion of 50 Bengalees. These were much lower than for Europeans accepted by Folin (1905). Campbell (1919) had also noted similar low excretion of sulphur in one Hindu, one Sikh and one Bengalee subjects in his studies in Singapore. Hence this present investigation.

METHODS

Urinary sulphur excretion of 50 healthy adults, staff and students of the Medical College, Baroda, was examined in months of March - July 1960. Their urine for 24 hours was collected in rubber corked glass bottles containing 2 ml of chloroform as preservative. The collection was noted regarding its volume, colour, specific gravity, reaction, deposits and abnormal findings, if any. The total inorganic sulphates were estimated by Folin's mothod using 5 per cent barium chloride solution as precipitant. The amount of ethereal sulphur was made out by difference between the above.

Since the excretion in this series also was low, a survey of the diet of the students together with the examination of the urine of these students was arranged. The diet survey was undertaken for 10 days by actually weighing the raw foods cooked in the mess and also taking into account, the extra food consumed by individual members and the cooked food wasted in the mess. The subjects whose diet was estimated also gave similar low excretory values of sulphur. Finding that their diet was adequate regarding total proteins, but poor in animal proteins, daily excretion of one subject whose diet included more animal proteins was scrutinized for a continuous period of one month.

To assess the relationship between S and N_2 excretion, total nitrogen was estimated by Kjeldahl's method and urea by hypobromite method in some cases.

The results are presented as studied and the usual way of reporting sulphur excretion in terms of SO_3 has been adopted here.

RESULTS AND DISCUSSION

Normal urinary sulphur excretion.— The chief source of sulphur in urine is the protein taken in diet. The sulphur liberated in the catabolism of dietary protein is largely converted to inorganic sulphates and absorbed. A small part of the inorganic sulphate thus derived and also from other sources is conjugated in the liver with products of bacterial decomposition and finally excreted as ethereal sulphates in urine. Most of the sulphur excreted in urine is in inorganic form 80 - 90 per cent the ethereal is only 6 - 8 per cent and these two - inorganic and ethereal vary with the protein intake of the subject. Thus Folin found 3.27 g of inorganic, 0.19 g ethereal and 0.18 g neutral sulphur as SO_3 excreted in 24 hours on protein liberal diet, whereas only 0 46 g inorganic, 0.10 g ethereal and 0.20 g neutral sulphur as SO_3 excreted on protein poor diet.

The neutral sulphur is said to be independant of the type of diet. It is hardly 4-5 per cent of the total sulphur and is excreted in fixed amounts of about 200 mg in healthy adults. Folin presumed it to represent, in classical terminology, the endogenous metabolism of proteins. In view of its little variability and confirmed independance of diet, estimation of this fraction was not taken up in our studies.

The findings in 50 normal adults in Gujarat are presented here.

TABLE I

Excretion of sulphur expressed as SO_3 in urine in 24 hours in normal adults

1	2	3	4	5	6
Name of worker	No. of subjects	Diet	Inorganic sulphur g	Ethereal sulphur g	Total (of 4+5) g
Present series	50 Gujarati	Veg.	Mean 0.860	0.111	0.971
			SD ±0.268	±0.079	±0.280
			SE Mean 0.038	0.011	0.039
Campbell (1919)	1 Hindu	Veg.	0.736	0.147	0.833
	1 Sikh	mixed	1.056	0.092	1.148
	1 Bengalee	mixed	0.829	0.102	0.931
Ray and Ganguli (1938)	50 Bengalee	mixed	0.650	0.100	0.750
Folin (1905)	European	Av. mixed	2.920	0.220	3.140
		Prot. liberal	3.270	0.190	3.460
		Prot. poor	0.460	0.100	0.560

The figures for sulphur excretion in Gujaratis are similar to those reported for other Indians. They are much lower than what are obtained for Europeans on mixed died. The ethereal sulphates are almost equal to and the inorganic only slightly more than those for Europeans on protein free diet. Folin reported an average excretion of 0.55 g of (inorganic + ethereal) SO_3 in 24 hours urine in subjects kept on protein free diet. This amount may be taken as derived from articles other than proteins. Why Indian subjects excrete sulphur near this basal level reported by Folin, remains to be examined.

TABLE II

Comparision of excretion of sulphur expressed as SO_3 in urine in 24 hours according to sex, age and economic state of the subjects

		No.of subjects	Inorganic sulphur g	Ethereal sulphur g	Total sulp. g
Sex	Male	40	Mean 0.884	0.112	0.996
			SD ± 0.288	± 0.084	± 0.296
			SE mean 0.045	0 013	0.042
	Female	10	Mean 0.764	0 093	0.857
			SD ± 0.140	± 0.062	+0.178
			SE mean 0.045	0.020	0.056
Age	20-30 yrs	26	Mean 0.898	0.106	1.004
			SD ± 0.137	± 0.082	± 0.320
			SE mean 0.027	0 016	0.063
	30-40 yrs	12	Mean 0.807	0.105	0.912
			SD ± 0.275	± 0.088	± 0.185
			SE mean 0.079	0 025	0.053
	40-50 yrs	12	Mean 0.814	0.122	0.026
			SD ± 0.240	± 0.058	± 0.293
			SE mean 0.069	0.017	0 085
Income	Rs. 100/-p.m.	9	Mean 0 806	0.059	0.865
			SD ± 0.292	±0.028	± 0.292
			SE mean 0.097	0.010	0.097
	Rs. 100-200 p.m.	23	Mean 0.770	0.104	± 0.874
			SD ± 0.146	± 0.079	± 0.190
			SE mean 0.031	0.017	0.044
	Rs. 200-300 p.m.	18	Mean 1.002	0.135	1.137
			SD ± 0.102	± 0.029	± 0.318
			SE mean 0 024	0.007	0.074
All Subjects		50	Mean 0.860	0.111	0.971
			SD ± 0.268	± 0.079	± 0.280
			SE mean 0.038	0.011	0.039

All the subjects reported by Ray and Ganguli (1938) and Campbell (1919) were males. The sex of the subjects seems to make little difference in sulphur excretion. In this series of 50 subjects, there were 40 males and 10 females. The average excretion for females was slightly but not significantly lower than for males.

Considering the excretion of sulphur in urine with reference to the age of subjects no significant difference was noticed in the various age groups.

Since the economic status of the family is likely to affect the nutritional status of an individual, this aspect was scrutinized. It revealed a slight increase in the excretion of sulphur in subjects of the upper economic group.

Excretion of sulphur and protein intake.—To examine therefore the relationship between food and sulphur excretion, a diet survey of the interne medical students hostel was undertaken for a continuous period of 10 days. Urine of these students was studied twice during the period of survey, regarding inorganic and ethereal sulphur, total nitrogen and urea. Their diet contained :

TABLE III

Average nutritional values per head per day of twelve subjects

Protein g	Fat g	Carbo. g	Calcium g	Phosph. g	Iron mg	Vitamin A i.u.	Vitamin B_1 i.u.	Vitamin C mg
77.4 7.4(a*)	94.5	397.3	0.76	1.76	33.12	1631	339.5	144.1

(a*) = Animal protein. Total calories = 2754.4

The same 12 subjects whose diet was surveyed showed urinary excretion values, during the same period, as follows :—

TABLE IV

Urinary excretion values in 24 hours collection of subjects (12) with estimated dietetic intake

	Volume in ml	Specific Gravity	Inorganic sulphur as SO_3 g	Ethereal sulphur as SO_3 g	Total Nitrogen g	Urea Nitrogen g
Mean	1452	1020.1	1.034	0.090	6.230	5.210
SD	± 540	± 5.1	± 0.350	± 0.053	± 1.487	± 1.780
SE mean	115	1.1	0.075	0.011	0.428	0.534

These subjects could be classed in the upper income group of the series. Their vegetarian diet was typical of middle class Gujarati food which has been proved to be inadequate regarding animal proteins. A diet survey of the middle class Gujarati families in Bombay by the Gujarat Research Society revealed 6.61 g of animal and 61.10 g of total protein intake per head per day. The M. S. University of Baroda conducted a diet survey of the middle class Gujarati families in Baroda and showed that per head per day consumption was (a) 4.75 g animal and 66.40 g of total proteins and (b) 7.74 g animal and 59.69 g total proteins by individuals with per capita income of (a) Rs. 25/- and (b) Rs. 25-50 per month respectively.

Thus, this vegetarian dietary, though not so inadequate in total protein content, is possibly made of such proteins which yield poor quantities of sulphur. The protein intake of the 12 subjects indicates a value of 2 873 g methionine per head per day. The other sulphur containing amino-acids as cystine is not estimated for want of information regarding the aminoacid composition of foods consumed by them.

Excretion of sulphur in one subject on better diet studied continuosly for a month. In order to ascertain whether sulphur was excreted in an erratic manner from day to day and also to note the effects of improved dietary, the sulphur excretion was studied in one subject (J. D. P.) continuously for a month. Although all his diet was not actually weighed, it included roughly a total protein content of over 70 g/day out of which atleast 30 g came from animal source. The methionine and cystine content of the animal protein (from milk and eggs) were found to be 1.21 g and 0.51 g respectively; thus totally equivalent to about 1.035 g of SO_3/day, He excreted 1.362 g SO_3 in 24 hours i. e. 50 per cent more sulphur in urine than the value for subjects studied here (Table 1). The figures of daily estimate indicated no erratic manner of day to day excretion either.

This proves that the characteristic low excretion of sulphur in Indians (Gujaratis) is not a racial peculiarity. Rationally thinking, it seems unlikely that the metabolism of Indians (Gujaratis) is peculiar in excreting abnormally low quantities of sulphur.

A similar study reported by Williams and Charles (1907) could be compared. Keeping on strict vegetarian diet, their European subject, in an experiment extended for 12 days, excreted an average of 0.811 g SO_3 in 24 hours. Thus, the studies in individual subjects conducted for a continuous period, hint that the urinary excretion of sulphur is predominantly governed by the type of food.

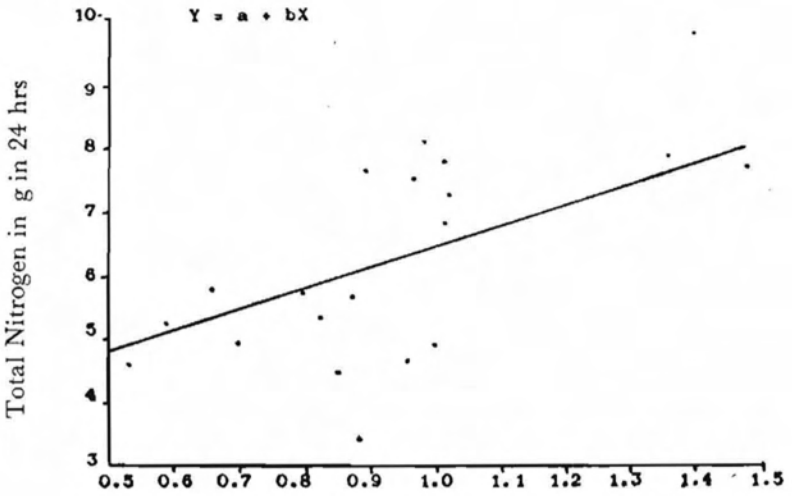
Urinary sulphur/nitrogen ratio.—Since excretion of nitrogen is accredited to be a true index of protein turn over in the body, the total nitrogen in urine in 24 hours was estimated in 23 cases together with its inorganic and ethereal sulphur content. The results are shown below :—

TABLE V

Comparision of urinary sulphur (as SO_3) and nitrogen value in 24 hours

Name of worker	Types of diet	Inorganic sulphur g	Ethereal sulphur g	Total nitrogen g
Present series	Veg.	mean 0.915	0.105	6.388
		SD ±0.322	± 0.074	± 0.580
		SE mean 0.067	0.015	0.329
Campbell (1919)	Veg.	0.736	0.147	5.970
Narayana (1935)	Veg.	.....	.....	6.200
	mixed	.....	.....	8.010
Ray & Ganguli (1938)	mixed	0.650	0.100	4.830
Gokhale (1939)		.....	.....	5.59
(1941)		.....	.....	6.09
Pai (1957)	Veg.	.....	.....	5.900
Folin (1905)				
	av. mixed	2.920	0.220	16.000
	prot. liberal	3.270	0.190	16.800
	prot. poor	0.460	0.100	3.600

Comparatively low nitrogen excretion has been reported by several workers - 6.20 g/24 hours on vegetarian diet and 8.01 g/24 hours on mixed diet by Narayana (1935); 4.83 g/24 hours on mixed diet by Ray and Ganguli (1938) and 5.90 g/24 hours on vegetarian Gujarati diet by Pai (1957). Since both sulphur and nitrogen are presumed to be derived chiefly from protein taken in food, excretion of sulphur and nitrogen should show some relationship. From this assumption some workers have accepted that "excretion of sulphur like that of urea or total nitrogen is an index of protein metabolism".



Inorganic + Ethereal SO_3 in g 24 hrs
Fig. 1

From this data, the relation between average (inorganic and ethereal) sulphur and nitrogen was statistically examined and a straight line $Y = a+bX$ could be fitted for the data very closely. The fitted line was found to be $Y = 2.919 + 3.424X$.

From the values of inorganic and ethereal sulphur/nitrogen, an apparant ratio of 1:15.7 is derived. Statistically examining the data the S:N ratio of 1:11 is not inconsistant and is acceptable. Examining Ray and Ganguli's data on Bengalees, a ratio of 1:16.1 for (inorganic and ethereal) sulphur/nitrogen is obtained. Wendt (1939) mentioned sulphur/nitrogen ratio of 1:14 in urine and 1:11 when total excretion in urine and stools were considered. This ratio cannot be so absolutely fixed and a variation within fair limits is expected dependant upon not only the total but also the type of protein consumed by the subject.

Other data.—Advantage was taken to study total volume, Sp.Gr., etc. in these samples. The results are tabulated on next page.

TABLE VI

Volume and specific gravity of urine excreted in 24 hours

		40 males	10 females	Total 50 subjects
Volume of urine in 24 hrs in ml.	Mean	1560.7	1059.0	1460.7
	SD ±	627.7	± 156.7	± 631.5
	SE mean	99.0	49.9	89.3
Sp. gr. of urine in 24 hrs.	Mean	1016.45	1016.85	1016.53
	SD ±	6.3	± 5.8	± 10.1
	SE mean	1.0	1.84	1.4

The studies were made during warm season in Baroda. The volume of urine is much large in 40 males than in 10 females. It may be due to their smaller built or perhaps their smaller intake of fluids. The Sp. Gr., however, gave a constant value of 1016 both in males and females. None of the subjects showed albuminurea, glycosuria or other abnormality. These figures fall within the range accepted as normal in physiology.

SUMMARY

Excretion of (inorganic + ethereal) sulphur expressed as SO_3 in urine in healthy Gujarati adult is on an average 0.9715 g in 24 hrs which is comparable with findings in Bengalees reported in literature, but is far lower than the values mentioned for Europeans living on mixed diet.

This low excretion of sulphur is attributed to the low protein-particularly low animal protein intake of these subjects. Simultaneous study conducted for a month on one subject whose diet included over 30 g of animal protein, indicated a notable increase in excretion of sulphur viz. 1.362 g SO_3 in 24 hrs.

An apparant S : N ratio in the present series is of the order of 1 : 15.7, but statistically considering a ratio of even 1 : 11 is admissible.

The average volume of urine excreted in 24 hrs by normal Gujarati adults is 1416.7 ml ± 631.5 ml which is just about the average accepted in texts. The Sp. Gr. of urine gave a fairly constant figure of about 1016 in 24 hrs' samples in warm months in which the studies were made.

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