Urinary 5-hydroxyindoleacetic acid in 8-hour collections as an aid in diagnosis of coeliac disease

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Challacombe, D. N., Goodall, M., Gaze, H., and Brown, G. A. (1975). *Archives of Disease in Childhood*, 50, 779. **Urinary 5-hydroxyindoleacetic acid in 8-hour collections as an aid in diagnosis of coeliac disease.** 8-hour urine excretions of 5-hydroxyindoleacetic acid (5-HIAA) were measured in 18 children with coeliac disease before treatment and the results expressed as μg 5-HIAA/mg creatinine. Similar measurements were made on urine collections from an age-matched control group of 24 children. Significantly higher values of 5-HIAA excretion were found in children with untreated coeliac disease. Measurement of the 5-HIAA: creatinine ratio in 8-hour urine collections is therefore proposed as an aid in the diagnosis of coeliac disease.

In an earlier communication (Challacombe *et al.*, 1972), the excretion of 5-hydroxyindoleacetic acid (5-HIAA) and creatinine was measured in 24-hour urine collections from children with untreated coeliac disease and from an age-matched control group. The results were expressed as μg 5-HIAA/mg creatinine. Significantly higher values were found in patients with untreated coeliac disease, when compared with the control group. After withdrawal of gluten from the diet, 5-HIAA excretion fell in all 8 cases who were followed up.

Urine collection over complete 24-hour periods from infants and young children presents technical problems which would be reduced or eliminated by collection over a shorter time interval. A further study has therefore been made on 5-HIAA excretion in timed 8-hour collection periods in a second group of coeliac patients and age-matched controls. The data from the study are presented in this report.

Materials and methods

Patients. Urine collections were made from 18 children with coeliac disease (Table I). Diagnosis was confirmed in each case by showing a flat duodenal mucosal surface in tissue obtained by peroral biopsy and by remission of clinical symptoms after introduction of a gluten-free diet. Control data were obtained from 24 children of approximately equal age (Table I), who were in hospital for treatment or assessment but did not have gastrointestinal disease.

Methods. All high serotonin-content foods (Ers-pamer, 1966) and all drugs were excluded from the diet for 24 hours before and during urine collection, which was made between 9.00 a.m. and 5.00 p.m. and passed directly into 10 ml glacial acetic acid. Collections were stored at -20°C until analysed. 5-HIAA was measured by a method based on that of Macfarlane *et al.* (1956), with improvements suggested by Mustala, Tuomisto, and Airaksinen (1964). Creatinine was measured by a standard Technicon AutoAnalyser alkaline picrate procedure (Methodology sheet N-11b) with a urine diluting step incorporated in the manifold.

Results

The 8-hour urinary 5-HIAA excretion values in patients with coeliac disease and the control group are shown in Fig. 1 and Table I. As in the previous study using 24-hour urine collections (Challacombe *et al.*, 1972), the values of the coeliac 5-HIAA: creatinine ratios in the 8-hour collections were higher than the control results. When considered as a homogeneous group overlap of data occurred, but this was almost entirely due to an age-related change in the ratio (Fig. 2). When coeliac results were compared with results from control children of similar age, only one was exceeded by a control value. Mean 5-HIAA: creatinine ratios for the coeliac and control groups were 23.2 and 9.3, respectively. Assessed by Student's 't' test (Table II), the difference between means for the two groups was highly significant ($P < 0.001$).

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TABLE I

Urinary 5-HIAA, creatinine, and 5-HIAA: creatinine ratios in controls and patients with coeliac disease

Case	Diagnosis	Age	Urine volume (ml)	5-HIAA (μ g)	Creatinine (mg)	Ratio
Controls						
1	Normal	3 w	61	163	10.4	15.7
2	Normal	1 m	41	108	7.4	14.6
3	Normal	2½ m	121	238	19.4	12.3
4	Normal	2½ m	216	421.2	23.8	17.7
5	Normal	3 m	173	437	31.1	14.0
6	Normal	7 m	180	458.6	30.6	15.0
7	Normal	9 m	85	500	28.9	17.3
8	Brain tumour	9 m	48	103	17.7	5.8
9	Normal	1 yr 3 m	330	550	95.7	5.7
10	Congenital heart disease	1 yr 8 m	87	162	26.9	6.0
11	Intussusception	1 yr 11 m	355	270.5	34.0	8.0
12	Tonsillitis	1 yr 11 m	60	347.1	50.0	7.0
13	Normal	2 yr 3 m	117	388	36.2	10.7
14	Sickle cell anaemia	2 yr 7 m	345	451.8	59.0	7.7
15	Normal	2 yr 9 m	13	52.4	4.5	11.6
16	Normal	3 yr 6 m	86.5	783	64.8	12.0
17	Normal	4 yr 3 m	175	610.8	134.7	4.5
18	Cerebral palsy	4 yr 8 m	85	665	75	8.9
19	Shigellosis	5 yr 3 m	215	996.3	133	7.5
20	Malaria	5 yr 4 m	1150	602.4	144	4.2
21	Normal	6 yr 1 m	187	750	136.5	5.5
22	Brain cyst	8 yr	96	267	99	2.7
23	Tracheo-oesophageal fistula	8 yr 3 m	125	448	94	4.8
24	Normal	8 yr 3 m	122	728	145	6.0
Coeliacs						
25		2 m	145	432	18.8	23.0
26		3 m	123	630	18.4	34.2
27		8 m	180	352.3	24.3	14.5
28		9 m	190	299.4	8.8	18.0
29		10 m	220	619.3	8.3	33.8
30		10 m	75	813.5	27.0	39.7
31		10 m	54	1001.2	26.0	38.5
32		1 yr	255	689.5	40.6	16.9
33		1 yr 1 m	59	847.8	27.7	30.6
34		1 yr 1 m	106	916.9	27.6	33.2
35		1 yr 3 m	18	378.9	13.7	27.7
36		1 yr 6 m	46	449.9	25.8	17.4
37		3 yr 2 m	290	813.7	49.3	16.5
38		3 yr 6 m	330	1110.0	66.0	16.8
39		4 yr 10 m	460	1739.0	106.0	16.4
40		5 yr 7 m	175	920.5	63.0	14.6
41		8 yr 5 m	310	1201.9	27.0	14.2
42		9 yr 6 m	215	2074.0	163.4	12.7

TABLE II

Statistical analysis of results (Student's 't' test)

	Volume (ml)	5-HIAA (μ g)	Creatinine (mg)	Ratio
Controls (n=24)				
Mean	186	437.5	62.5	9.3
SD	(225.7)	(243.8)	(48)	(4.5)
Coeliac disease (n=18)				
Mean	180	849.4	41.2	23.2
SD	(117)	(469)	(38)	(9)
Comparison of means				
t	0.1	3.39	1.5	5.8
P	>0.9	<0.003	>0.13	<0.001

Mean 8-hour urine volumes and their creatinine content were similar for the two groups. The differences between means were not significant. In both 8-hour and 24-hour collections of urine (Challacombe *et al.*, 1972), the values of the ratio in both controls and coeliacs were shown to be high in early infancy and fell to lower values at approximately one year. They then appeared to fall more slowly over the remaining age range investigated. The ratios in the 8-hour collections in this control group tended to be lower than those in the 24-hour control collections. Ratios in the 8-hour and 24-hour collections in patients with coeliac disease were, however, equally high.

FIG. 1. — 5-HIAA:creatinine ratios in 8-hour urine collections from children with untreated coeliac disease (●) and age-matched controls (○).

FIG. 2. — 5-HIAA:creatinine ratios in patients with untreated coeliac disease and controls, unrelated to age.

Discussion

Estimation of urinary excretion of 5-HIAA, expressed as a 5-HIAA:creatinine ratio, can be used as an aid in the diagnosis of coeliac disease using urine collections made over periods as short as 8 hours. Since the definitive diagnosis of coeliac disease can only be made by peroral small intestinal biopsy, 5-HIAA excretion may be used as a further indication in patients who do not have all

the classical symptoms of coeliac disease but in whom the diagnosis is suspected.

Urine collections were made between 9.00 a.m. and 5.00 p.m., that being the period when the collection can be supervised most easily, either in the ward or at home. The period of collection must be consistent from patient to patient to avoid the influence of diurnal variation in 5-HIAA excretion which has been reported (Erspamer, 1966). The difference between 8-hour and 24-hour 5-HIAA:creatinine ratios seen in the control data may be a consequence of diurnal variation of 5-HIAA excretion. This difference was not seen in the coeliac patients, suggesting that excretion is not only abnormally high, but that the diurnal excretion pattern is disturbed.

Factors that may be responsible for increased excretion of 5-HIAA in the urine in children with untreated coeliac disease have been discussed previously (Challacombe *et al.*, 1972). An abnormality of argentaffin cell metabolism was proposed which may be stimulated or shown by the introduction of dietary gluten, producing increased quantities of 5-hydroxytryptamine and its metabolite 5-HIAA. Measurement of 5-HIAA excretion on children who have sibs with this disorder before the introduction of gluten into their diet may throw further light on this hypothesis.

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