The Value of Estimating Urinary Indican

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SUMMARY

Urinary indican, a metabolic product of bacterial action on tryptophan in the small bowel, has been estimated in the urine as an indirect indication of small bowel bacterial contamination, in 50 normal controls, 18 patients with small bowel disease, 22 patients with calcific pancreatitis and group of 20 children with kwashiorkor and 10 control children of a similar age-group.

Elevated urinary indican excretion 2SD above the mean normal of 49 mg/24 hours was found in a variety of small bowel conditions. Patients with calcific pancreatitis had a mean indican level of 48 mg/24 hours, suggesting that there is no excess bacterial proliferation in the small bowel in this disease.

Urinary indican levels were extremely low in both control children and kwashiorkor patients. An insignificant increase occurred after the kwashiorkor was treated. The value of estimating the indicanuria level in each group is evaluated.

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There has been a renewed interest in the bacterial flora of the bowel with the recognition that altered bowel anatomy or motility can lead to bacterial proliferation in the small bowel with subsequent metabolic disturbances. These include vitamin B_{12} malabsorption and increased deconjugation of bile salts to high concentrations of bile acids.

Since diagnostic jejunal and ileal intubation to study the bacterial flora is time-consuming and the bacteriological methods are complex and imperfect, screening tests have been devised for use in clinical practice to establish the presence of an increase in small bowel flora. These include tests of vitamin B_{12} absorption which remains abnormal in the presence of intrinsic factor but improves with antibiotics; steatorrhoea, as measured by faecal fat, which improves with antibiotics; and an increased urinary excretion of indican which can be reduced by the administration of antibiotics.[1-4] An increase in the urinary excretion of indolic compounds in rats with intestinal pouches was first reported by Donaldson *et al.*[5] in 1961 and shown subsequently to be due to abnormal intestinal bacterial activity on tryptophan. Tryptophan is converted to indole by the metabolic activity of intestinal bacteria. The bacteria responsible for this are indole-producing micro-organisms such as *E. coli*, bacteriodes and some strains of klebsiella. The indole is then hydroxylated in the liver to 3-hydroindole or indoxyl, and this in turn is conjugated with sulphate to indoxyl sulphate or indican which is excreted in the urine (Fig. 1).

There are numerous gastro-intestinal abnormalities associated with increased bacterial flora in the small bowel (Table I).[6] Normal gastric acid secretion probably limits the entry of bacteria into the small bowel and therefore gastric abnormalities such as afferent loop stasis and achlorhydria constitute one group. A second group includes small bowel abnormalities with areas of statis due to strictures, blind-loops, diverticulae, or where the auto-

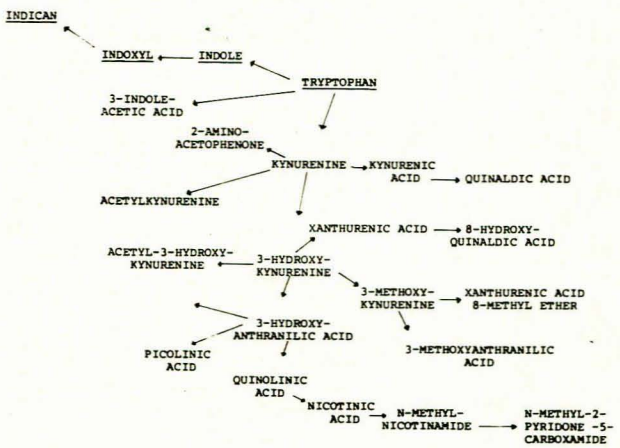

Fig. 1. Tryptophan metabolism.

TABLE I. CONDITIONS ASSOCIATED WITH INCREASED SMALL BOWEL BACTERIAL FLORA

Gastric abnormalities:
- **(a) Afferent loop stasis**
- **(b) Malfunctioning gastrojejunostomy**
- **(c) Achlorhydria**

Small bowel abnormalities:
- **(a) Strictures — Congenital**
  - **Crohn's disease**
  - **Tuberculosis**
  - **Lymphoma**
  - **Postirradiation**
- **(b) Surgical blind-loops**
- **(c) Small bowel diverticulosis**
- **(d) Intestinal obstruction and pseudo-obstruction**
- **(e) Abnormal autonomic innervation of the bowel**
  - **Scleroderma**
  - **Diabetic neuropathy**
  - **Postvagotomy**

Miscellaneous abnormalities:
- **(a) Ileocolic anastomosis**
- **(b) Gastrocolic fistula**
- **(c) Enterocolic fistula**
- **(d) Massive intestinal resections**
- **(e) Partial biliary obstruction with cholangitis**

nomic innervation of the bowel is impaired as in scleroderma or diabetic neuropathy. Thirdly, there may be free communication between the small bowel and colon as with an ileocolic anastomosis, gastrocolic fistula, enterocolic fistula or following massive intestinal resections. Under these conditions the small bowel is exposed to high concentrations of colonic bacteria.

Enteric infection is common in children with kwashiorkor[7] and low indican levels have been reported in the few patients with pancreatic steatorrhoea investigated.[1,3,4]

The present paper reports our findings on urinary indican levels in normal adult and infant controls, in certain malabsorptive states and compares it with results found in pancreatic insufficiency and kwashiorkor. The value of estimating the indicanuria levels in each group is evaluated.

MATERIAL AND METHODS

Five groups of patients were investigated;

1. Fifty normal adult controls comprising 7 medical students, 11 nurses and 32 non-gastro-intestinal ward patients, none of whom was on antibiotic treatment.

2. Eighteen patients with a variety of malabsorption states, comprising 3 with Crohn's disease, 3 with post-gastrectomy afferent loop syndrome and steatorrhoea, 2 with coeliac disease, 2 with malabsorption of unproved cause, 2 with ileocolic anastomoses and 1 each of the following: small bowel diverticuli, idiopathic small bowel dilatation after total gastrectomy, subacute intestinal obstruction and short-bowel syndrome. In 6 of these patients, indican levels were measured before and after therapy with antibiotics.

3. Twenty-two patients with calcific pancreatitis, of whom 15 had steatorrhoea; all had a grossly abnormal pancreatic function test after secretin-pancreozymin stimulation.[8]

4. Twenty children with overt kwashiorkor.

5. Ten control children of a similar age to the kwashiorkor patients.

A 24-hour urine collection was made into a bottle containing 10 ml of chloroform. An aliquot of urine was taken and frozen at −20°C until the estimations were performed. Urine indican levels were determined in duplicate using the method of Curzon and Walsh.[9]

RESULTS

The mean urinary indican level in the 50 adult controls was 49 mg/24 hours ± 24 mg. A wide range was found in normal controls from 7 mg/24 hours to 103 mg/24 hours (Fig. 2). In patients with malabsorption states, the mean indican level was 144 mg/24 hours ± 120 mg and in this group, the patients with suspected bacterial overgrowth as evidenced by a response to tetracycline or overt intestinal stasis radiologically, the mean level was 185 mg/24 hours ± 130 (Figs. 2 and 3). In 6 cases in which the indican level was measured before and after tetracycline therapy, the mean indican levels were 178·3 mg/24 hours and 83·5 mg respectively.

The value of estimating urinary indican levels is well illustrated in a case with an ileal bypass in whom ileocolic anastomosis was carried out following an episode of intestinal obstruction. Following the operation, the patient developed

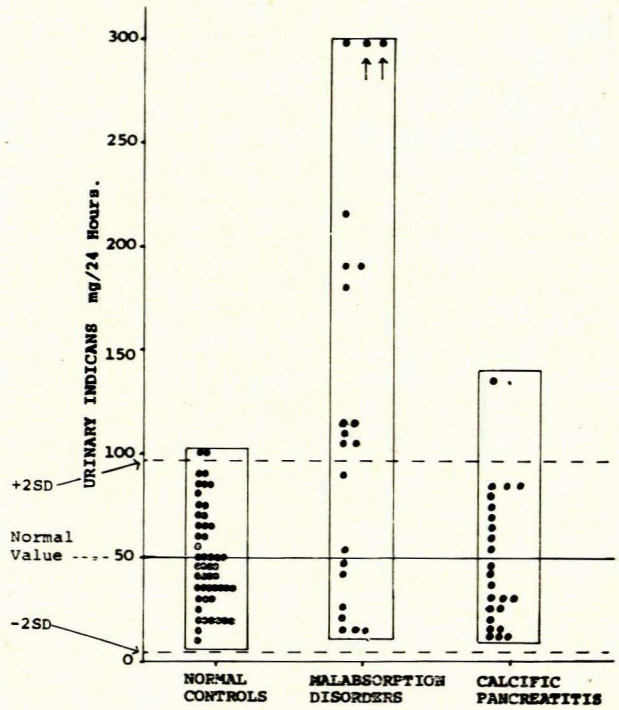

Fig. 2. Urinary indican excretion in normal controls, patients with a variety of malabsorption disorders and calcific pancreatitis.

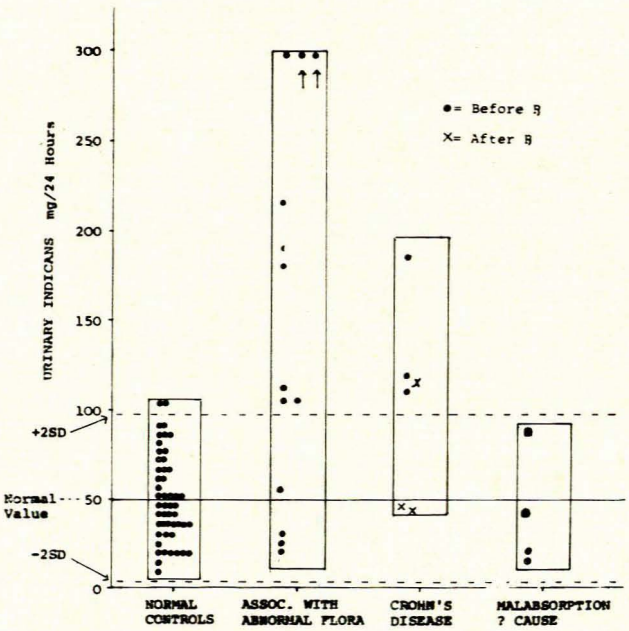

Fig. 3. Urinary indican levels in normal controls, cases of potential bacterial contamination and other malabsorptive states and Crohn's disease.

persistent diarrhoea and steatorrhoea. Investigations confirmed a mild steatorrhoea with a normal jejunal biopsy. Urinary indican levels measured on 2 days before and after tetracycline therapy were 393 and 330 mg/24 hours and 157 and 136 mg/24 hours respectively. The patient then underwent surgery to re-establish normal continuity of the bowel. The urinary indican levels fell to 20 and 39 mg/24 hours. The levels correlated well with the improvement of vitamin B_{12} absorption from 8% to 28% (Fig. 4).

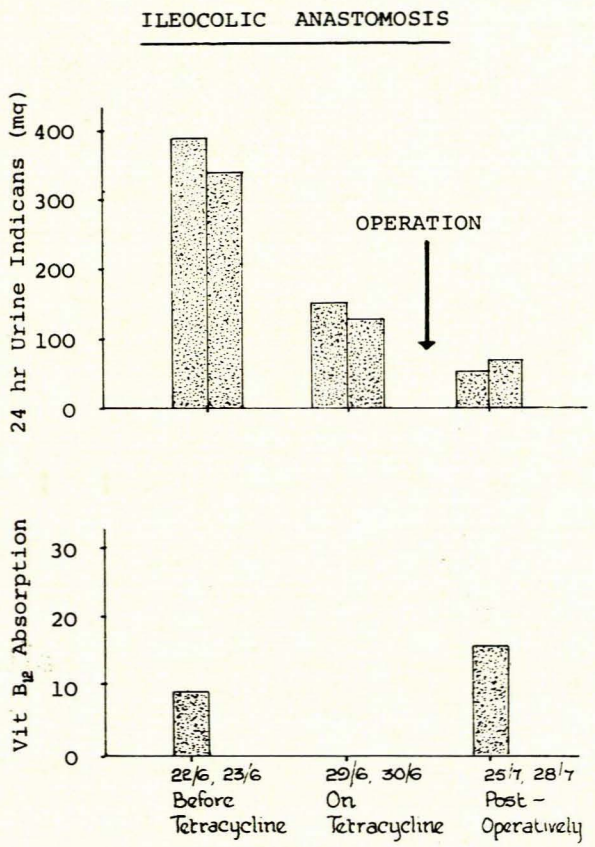

Fig. 4. Response of indican levels and vitamin B^{12} absorption in a patient with an ileocolic anastomosis to tetracycline and surgery.

The 22 patients with calcific pancreatitis had urinary indican levels similar to the control adult group with a mean of 48 mg/24 hours ± 32 mg (Fig. 2). There did not appear to be any correlation between the indican levels and the severity of the steatorrhoea. One patient with pancreatic steatorrhoea had a high indican level (>100 mg/24 hours).

Urinary indican levels in children were considerably lower than in adults. The mean urinary indican level in the 10 control children was 13 mg/24 hours ± 7 mg (Fig. 5). In the 20 children with kwashiorkor on admission and before the institution of therapy the mean level was 9 mg/24 hours ± 6 mg. This difference was not statistically significant (P>0·05). The level of indicanuria was re-estimated in 11 of these children after treatment of the kwashiorkor. The mean level then was 15 mg/24 hours. The increase above pretreatment levels was not significant.

In 4 of the children with kwashiorkor, the indican levels were re-estimated after feeding with 2·5 - 5 g of tryptophan. In 2 cases the levels remained the same, while in the other 2 cases levels increased from 16 and 9 mg/24 hours to 124 and 75 mg/24 hours respectively. Preliminary studies by small bowel intubation and bacterial culture done on 2 patients showed high bacterial colony counts of aerobic coliforms consisting of *E. coli,* klebsiella and proteus to levels of $4{\cdot}2 \times 10^{11}$ and $2{\cdot}8 \times 10^{6}$.

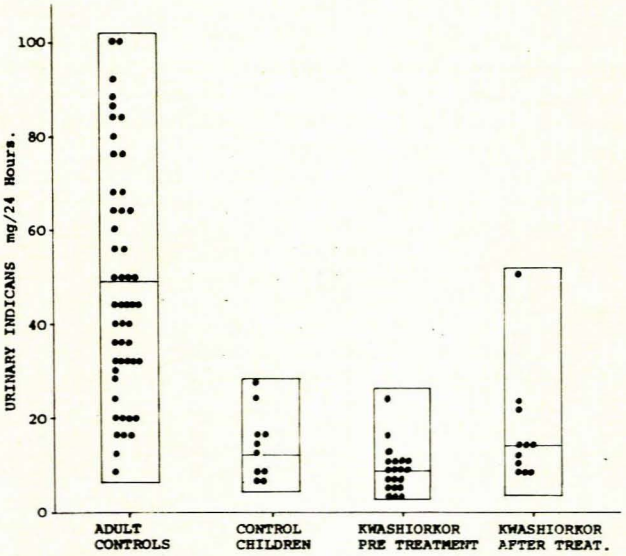

Fig. 5. Urinary indican levels in adult and children controls, and children with kwashiorkor before and after treatment.

DISCUSSION

This study has confirmed the finding of other workers[1-3] with regard to both the level of urinary indican excretion in normal adult controls and patients with small bowel disease. Raised levels (>100 mg/24 hours) were found in patients with a variety of malabsorptive states. Fordtran *et al.*[10] showed that raised indican levels could occur in cases of small bowel malabsorption without a demonstrable increase in small bowel bacteria and they suggested that this is due to colonic bacterial action on unabsorbed tryptophan with subsequent absorption of indoles in the colon. In the group of patients with conditions associated with bacterial overgrowth in the small bowel, the finding of high urinary indican levels (>200 mg/24 hours) is virtually synonymous with bacterial overgrowth and these patients can be expected to respond to antibiotic therapy or surgical correction of an anatomical abnormality. However, the finding of a normal indican level does not absolutely exclude the above diagnosis.

On the other hand we were unable to demonstrate abnormal indicanuria in patients with overt steatorrhoea due to pancreatic insufficiency. This indicates that these patients have no appreciable bacterial overgrowth of indole-producing organisms or any significant malabsorp-

tion of tryptophan in the small bowel despite protein maldigestion. Although abnormalities in small bowel function tests do occur sporadically in patients with pancreatic steatorrhoea[11,12] the cause is unlikely to be due to bacterial contamination of the bowel.

One can only speculate on the low indican levels in children compared with adults. It is likely that the bacterial flora of the small bowel in children has lower concentrations of indole-producing bacteria and high concentrations of lactobacilli. The even lower levels of indicanuria in the kwashiorkor patients might well be due to a lack of available protein and thus of the amino acid tryptophan necessary to be converted to indole by the reported increased bacterial population of coliforms in the small bowel. The finding of low levels of essential plasma amino acids in children with kwashiorkor and high levels following dietary therapy[13] would tend to support this, as would the effect of tryptophan feeding in the 4 children reported. Estimation of urinary indican is thus of no value in indicating whether bacterial overgrowth occurs in the small bowel in children with kwashiorkor, as the levels are invariably low.

Estimation of indican in the urine in cases of possible small bowel bacterial overgrowth both before and after treatment may give a valuable additional parameter to the diagnosis and assessment of therapy. The method is fairly simple and cheap and is not unpleasant to the patient, as is small bowel intubation, and may be carried out in any hospital with reasonable laboratory facilities.

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