

Title: Gastrointestinal Studies in Normal Thais

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**OBJECTIVE:** Studies of small bowel histology, bacteriology and function were carried out in asymptomatic Thais in order to provide baseline data for future studies of diarrheal disease.

**Description:** These studies were carried out on 34 adult Thai villagers from the Bangkok suburbs. None gave a history of recent gastrointestinal complaints. Several were anemic, and a number had one or more intestinal parasites. Each volunteer was hospitalized for a general workup and the gastrointestinal function tests listed in Table I. As the table indicates, some of the tolerance tests were repeated at times with different dosages of test substance. Small bowel biopsy was performed with either a Crosby-Kugler or Carey biopsy capsule. At the time of biopsy, small bowel juice was obtained either by siphonage through the biopsy capsule or by aspiration through a second tube taped to the biopsy capsule tubing. Part of the tissue was fixed in 10 per cent buffered formalin for dissecting microscope studies and histologic examination. Two other portions were immediately frozen in dry ice for histochemical studies and mucosal disaccharidase assays. The specimens for disaccharidase assay were stored for up to 2 weeks at  $-70^{\circ}\text{C}$ , then were shipped in dry ice to Dr. Pearl Anderson of WRAIR, who performed the assays.

Less extensive studies were carried out on 30 children in an orphanage whose ages ranged approximately from  $2\frac{1}{2}$  to  $9\frac{1}{2}$  years. After a general medical examination and laboratory evaluation, lactose, xylose and glucose tolerance tests were performed. Small bowel biopsy was performed using a pediatric-size Crosby-Kugler biopsy capsule, and intestinal juice for bacteriologic studies was obtained as in the adult subjects. The specimens were smaller than those obtained from adults, so that tissue was not available for histochemical examination but histologic examination and disaccharidase assays were done.

**RESULTS:** Table I summarizes the data obtained on the 34 adult patients whose studies have been completed. Some of the volunteers were unable to stay in the hospital long enough to have every test performed, but biopsies, lactose, and xylose tests were done in nearly all subjects.

The 5-hour urinary excretion of xylose averaged  $23.6\% \pm 6.5$  (S.D.). This compares well with Sheehy's published normal values of  $22.8\% \pm 5.6$  in 114 U.S. normal. Xylose determinations were done on blood drawn 2 hours after the administration of the sugar in 19 of the subjects. The concentrations ranged between 29 and 53 mg/100 ml, with an average of 42 mg/100 ml. Two subjects had diarrhea after administration of the sugar, in these the 5-hour excretion was less than 15 per cent. No evidence of renal or hepatic disease which might affect xylose metabolism, was found in any subject.

Lactose tolerance test were abnormal in 33 of 34 subjects tested. The single normal response, a maximum rise of blood glucose of 41 mg/100 ml, was found in a woman in the 8th month of pregnancy. The test was repeated 3 months after delivery and showed a maximum blood glucose rise of 2 mg/100 ml. Repeat tests in 7 other subjects remained abnormal. These data correlate well with the results of lactase assays of jejunal tissue (see below). Diarrhea occurred in 19 of the 34 subjects, and was usually accompanied by mild cramps.

Glucose tolerance tests were performed in 23 subjects using a dose of 0.75 gm/Kg body weight, an amount equimolar to the glucose delivered in the standard dose of lactose of 1.5 gm/Kg body weight. Only 3 subjects demonstrated a blood sugar rise of less than 20 mg%. The test was repeated in 2 of these 3 subjects using a 75 gm dose, and 1 responded normally.

Co<sup>57</sup>-labelled vitamin B<sub>12</sub> was administered together with Intrinsic Factor to 23 patients and to an additional patient without Intrinsic Factor. Absorption was normal in all subjects.

Serum beta-carotene concentrations were measured in 28 subjects, and was found to range between 54 and 336 mcg/100 ml with an average of 153 mcg/100 ml. These values are all within normal limits.

Vitamin A tolerance tests were performed in 20 subjects using a dose of 250,000 IU of the vitamin in corn oil, with serum sampling at 0, 3, and 5 hours. The concentration of the vitamin in serum failed to rise by 200 per cent in 4 subjects, 1 of whom was found to have abnormal excretion of fat in the stool. One patient also had a low-normal xylose tolerance test, but a fat tolerance test was not done.

Fecal fat excretion was less than 5 gm per day in all but 1 of the 17 subjects whose diets were supplemented with 75 gm of butter/day. The one abnormal patient excreted an average of 6.8 gm per day on this diet.

Data on mucosal disaccharidase assays (Table II) have been completed on 12 subjects to date. Activity is referred either to wet weight of tissue or to tissue protein content. Lactase activity per gm protein was low in 7 subjects, but was low in all 12 when expressed in terms of tissue weight. Sucrase activity was low in 3 subjects by either criterion, and maltase activity was abnormal in 1 patient by both criteria. The normal lactase values per gram of protein were, however, barely normal, ranging from 4.6 to 7.7 units/gm tissue. The normal range in the laboratory doing the determinations is 4-149, with a mean of 49 units/gm tissue. Change in the water content of the tissue due to storage in the frozen state may explain the discrepancy noted in lactase results between the two methods of expression of the data. Calculations of protein content from normal U.S. data provided by Dr. Anderson gave an average value of 0.066 gm protein/gm wet weight, whereas the average value for the 12 patients was 0.085 gm protein/gm wet weight. Only 3 of the 12 patients had protein concentrations below the average of Dr. Anderson's laboratory. Whether this difference in composition is real or related to storage of the tissue prior to assay is unknown.

The radiologic appearance of the small bowel was normal in all 11 Subjects examined with the exception of an increased transit time in 1 subject and evidence of ascariasis in 2 subjects.

Detailed pathologic studies of the mucosal biopsies have not been completed, but several trends are apparent. Abnormal appearance of the mucosa under the dissecting microscope was seen frequently. The majority of the villi were leaf-shaped rather than finger-shaped. In some specimens true villi were not seen but ridges and convolutions were present instead. The histologic changes consisted of mild infiltration of the lamina propria, primarily with mononuclear cells, in nearly all the biopsies. Occasional sections showed mononuclear cells migrating through the epithelium. The ratio of villus height to crypt depth was normal. Histochemical studies were begun just prior to the end of this reporting period, and too few patients have been studied to justify comment.

The results of the absorption studies in the 30 children under study are summarized in Table III. Only 1 child, a 9 year old boy, had a blood glucose rise of over 20 mg/100 ml after a dose of lactose of 2.0 gm/Kg. This child had entered the orphanage only 6 months before, and had consumed large amounts of milk almost all his life. All children in this institution receive 8 oz of milk daily, but the history of milk intake prior to admission is generally unknown. All the children had normal glucose tolerance tests, while half had abnormal xylose tolerance tests.

Pathologic examination of the pediatric biopsies is incomplete. The specimens examined to date show occasional branched villi, some leaf-shaped villi, and a definite increase in the distance between villi. These changes are less pronounced than in the adult biopsies. Histologic studies have been completed on so few biopsies that comment on this aspect of the study must be deferred.

DISCUSSION: The adults studied to date have been characterized by evidence of a mild, nonspecific jejunitis without villous atrophy and a fairly discrete loss of the ability to absorb lactose.

The high prevalence of mild nonspecific jejunitis in biopsies from Thailand was previously noted by Sprinz and co-workers. In the present study an inflammatory infiltrate was the only abnormality found. The shortening and thickening of villi, deepening of the crypts, and other changes suggestive of sprue noted by Sprinz were either absent or minimal. Likewise, the intestinal function tests (with the exception of lactose tolerance) were normal. In the previous study xylose excretion was severely impaired in 13 per cent of the persons studied, while in the present study the xylose excretion was not significantly different from 1114 normal patients in the U.S. reported by Sheehy. The reasons for these differences are not clear.

The available evidence suggests that the abnormality of lactose absorption is due to deficiency of lactase. The studies on Children suggest that this defect appears early in life. It may be related to dietary habits, for few Thais drink milk after weaning. It is possible that the lack of enzyme activity is due to absence of substrate in the diet. A second possibility is that this is a reflection of injury to the mucosa. This is supported by the mild inflammatory changes seen in nearly every biopsy. A study planned for the next quarter will attempt to induce lactase activity in adults by the administration of the sugar. In addition, the effect of measures such as eradication of parasites, correction of anemia, and the administration of folic acid or tetracycline, which are beneficial in tropical sprue, may further clarify the significance of these findings,

Table I

Test	Dose	Normal Value	Subjects	Abnormal
Xylose Tolerance	25 gm or 0.5 gm/Kg	23%/5 hours	26 } 2 }	0
	5 gm	+ 5.6 (1S. D.)	5 }	0
Lactose Tolerance	1.5 gm/Kg	20 mg% rise	32 } 2 }	33
	75 gm repeat 1.5 gm/Kg		8 }	8
Glucose Tolerance	0.75 gm/Kg	20 mg% rise	23	3
Vitamin B <sub>12</sub> Absorption	—	7.0%/24 hours	24	0
Vitamin A tolerance	260,000 U	200%/ rise	20	4
Serum carotene	—	> 40 mcg/100 ml	28	0
Fat tolerance	75 gm/day	< 5 gm/day	17	1

Table II

Jejunal Disaccharidase Activity: Number Abnormal in a group of 12 subjects.

Enzyme	U/G Tissue	U/G Protein
Lactase	12	7
Maltase	1	1
Sucrase	3	3

Table III

Test	Dose	Normal Value	Subjects	Abnormal
Lactose tolerance	2 mg/Kg	> 20 mg% rise	32	31
Xylose tolerance	0.5 gm/Kg	> 10%/5 hrs	24	12
	5 gm	> 10%/5 hrs	23	11
Glucose tolerance	1 gm/Kg	> 20 mg% rise	32	0

Age range 23/4 — 91/2 Years