SEROTONIN AND THE GASTROINTESTINAL TRACT*

BERNARD J. HAVERBACK, M.D., AND JOHN D. DAVIDSON, M.D.

From the Clinic of General Medicine and Experimental Therapeutics, National Heart Institute, National Institutes of Health, Public Health Service, U. S. Department of Health, Education and Welfare, Bethesda, Maryland, and the University of Southern California School of Medicine, Los Angeles, California

The major body depot of the indole amine, serotonin, is the gastrointestinal mucosa. Serotonin is localized to the enterochromaffin cells of the mucosa which extend from the esophagocardiac junction to the anus.[1] Minor depots include the brain and the blood platelets. The latter serve as a vehicle for this amine, with no detectable serotonin being found in the plasma normally. Serotonin is largely metabolized by monoamine oxidase to 5-hydroxyindoleacetic acid (5-HIAA) which is excreted in the urine.[2]

The function of the serotonin in the enterochromaffin cells of the gastrointestinal tract remains obscure. Previous studies have demonstrated that serotonin and its precursor, 5-hydroxytryptophan (5-HTP), inhibit acid secretion but stimulate mucus secretion of the stomach.[3] Also, observations have shown that the amine is a stimulus to intestinal motility.[4, 5] The anatomic proximity of the enterochromaffin cells to the terminal fibers of the submucous plexus was first described by Masson,[6] who thought that these cells have a neuroendocrine role in the intestinal tract. In the light of these facts it is probable that serotonin, elaborated by the enterochromaffin cells, is important in the regulation of intestinal motility.

Studies of the effects of serotonin administered intravenously in the gastrointestinal tract *in vivo* have been limited by the cardiovascular and pulmonary responses to the amine and by its rapid degradation to its inactive metabolite, 5-HIAA. Also, the effect of intracellular serotonin may be different from that of extracellular serotonin. Administration of the serotonin precursor, 5-HTP, results in an increase in intracellular as well as extracellular serotonin.[7, 8]

As serotonin has potent pharmacologic effects on the gut, the status of this naturally occurring amine in certain intestinal diseases has remained a point of interest.

The purpose of this study was to (1) determine the effect on intestinal motility and intraluminal pressure in man of endogenously produced increases of serotonin by administration of 5-HTP; (2) evaluate by studies in an unusual case the contribution of the human intestinal tract to blood serotonin and urinary 5-HIAA levels; and (3) present preliminary studies of blood serotonin and urinary 5-HIAA levels in a small group of patients with intestinal disorders. In the course of these studies, 1 patient was discovered to have a high urinary excretion of another indolic metabolite, indole-3-acetic acid.

Patients and Methods

The effect of 5-HTP and other drugs on motility and intraluminal pressure in the jejunum was determined in 7 adults who were free of gastrointestinal disease and in 2 patients with the nontropical sprue syndrome. The recordings were made over several hours while the subjects were in a supine position. The measuring apparatus consisted of a P23 B Statham electrical transducer, a bleeder system, and a Sanborn Polyviso amplifying recording unit, as described by Lorber and Shay.[9] The recording sensitivity was such that a 10-mm. pen deflection was equivalent to 5 cm. of water pressure. The paper speed was 0.5 mm. per second. The following substances were administered intravenously: 25 to 50 mg. of 5-HTP dissolved in isotonic saline was given by means of a constant infusion pump over a period of 30 minutes; serotonin creatinine sulfate was infused at rates varying from 0.3 to 1.2 mg. per minute; atropine sulfate in a dose of 1 mg. was given in a single injection. As indicated below, more than one agent was used during a single experiment. Two subjects free of gastrointestinal disease were pretreated with the serotonin antagonist, *brom*-lysergic acid diethylamide (BOL),

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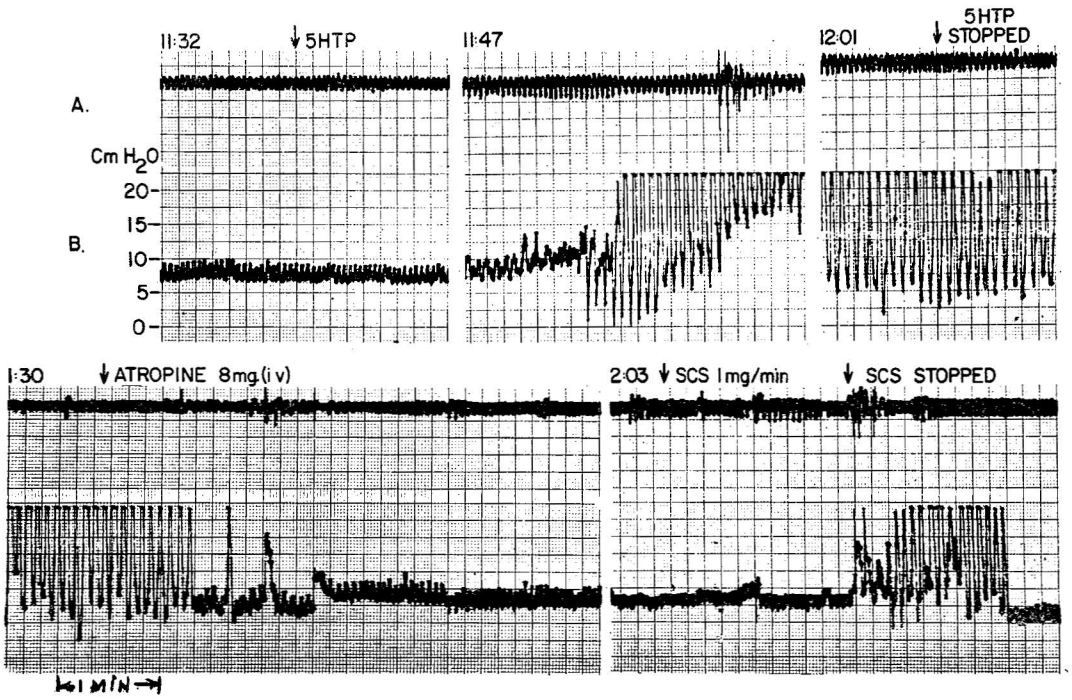

FIG. 1. Respiration (*tracing A*) and jejunal motility (*tracing B*) in a control subject. Following administration of 5-hydroxytryptophan (5-HTP) in a dose of 40 mg., a marked and sustained stimulus to motility occurred. Atropine sulfate inhibited motility stimulated by 5-HTP but not that stimulated by serotonin.

in a dose of 2 mg. orally prior to administration of 5-HTP.

Platelet serotonin and urinary 5-HIAA were measured by the method of Udenfriend *et. al.*[10] A patient (F. W.) whose intestinal tract had been almost entirely resected was studied to learn more about the contribution of the gut to the levels of these substances in blood and urine. The patient was a 39-year-old woman whose entire colon, except for a small segment of rectum, had been excised because of multiple polyposis. Subsequently, most of the small intestine had been resected because of an infiltrating desmoid tumor of the anterior abdominal wall, with anastomosis of the proximal jejunum to the rectal segment. To investigate serotonin metabolism in patients with intestinal disorders multiple determinations of blood serotonin and urinary 5-HIAA were made in 2 patients with nontropical sprue (T. D. and R. A.), in 2 with ulcerative colitis (F. B. and C. R.), in 2 with regional enteritis (J. R. and N. W.), and 1 with Whipple's intestinal lipodystrophy (T. E.).

Results

Within 6 to 40 minutes after the start of 5-HTP infusion a marked increase in intestinal motility was observed in each of the normal subjects (fig. 1). Usually there was no marked increase in the intraluminal pressure, but in a few instances increases up to 15 cm. of water were noted and abdominal cramping was experienced. The pressure of the motility waves increased up to approximately 25 to 35 cm. of H_2O. The increased motility persisted for at least 90 minutes following the start of the 5-HTP infusion. No effect on pulse, blood pressure, respiration, or mental status was noted following the dose of 5-HTP used. In 3 subjects 1 mg. of atropine sulfate was administered during the stimulated phase of intestinal motility, and in each study inhibition of intestinal motility was almost complete. However, when serotonin creatinine sulfate was administered at this point, the stimulus to intestinal motility was not modified (fig. 1). In 1 patient with nontropical sprue the response of

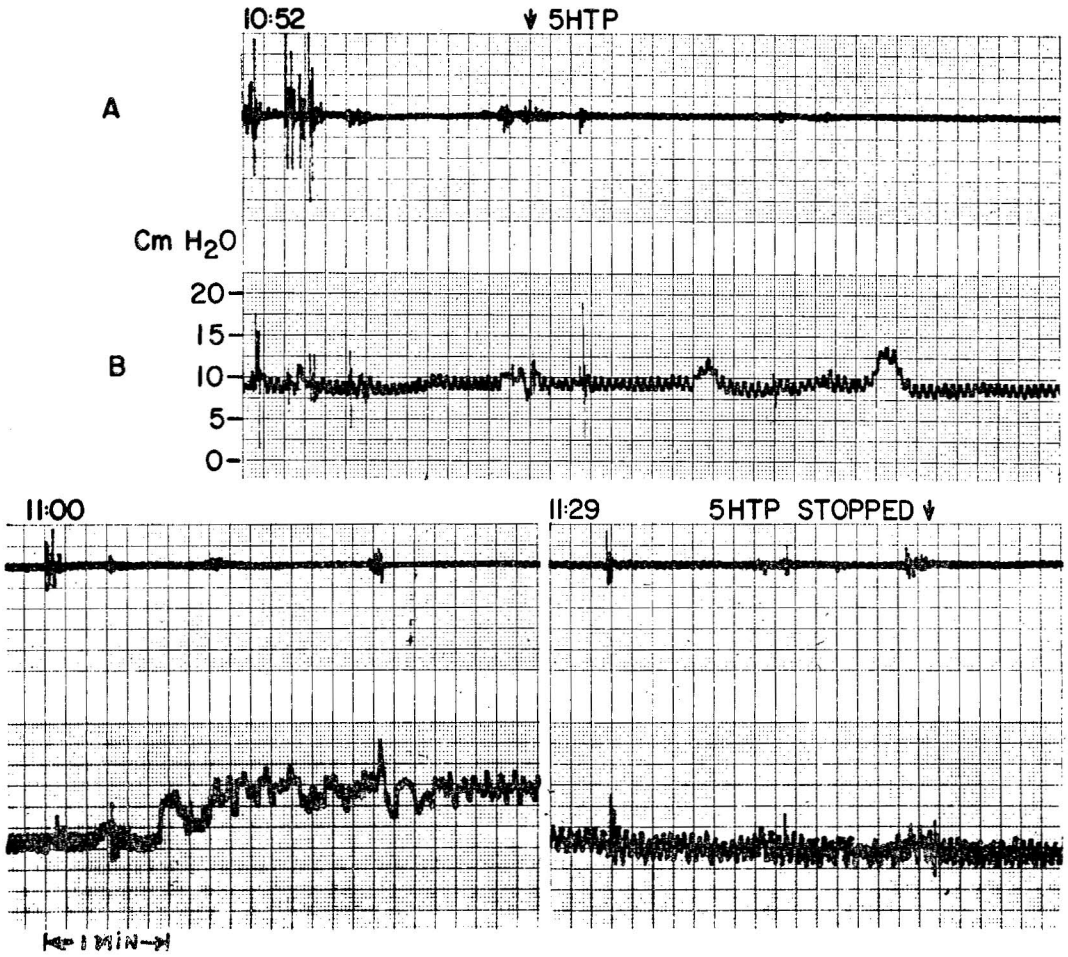

FIG. 2. Respiration (*tracing A*) and jejunal motility (*tracing B*) in a patient with nontropical sprue. Following administration of 5-hydroxytryptophan (5-HTP) in a dose of 40 mg., the resulting stimulus to intestinal motility was much smaller and less prolonged than that in the control subject.

the intestine to 5-HTP, atropine, and serotonin creatinine sulfate was identical to that in the normal. However, in the other patient with nontropical sprue the intestinal response to 5-HTP was markedly diminished (fig. 2).

When BOL was administered to a dog in a dose of 4 mg., intestinal motility stimulated by serotonin was effectively inhibited (fig. 3). Following administration of 2 mg. of BOL, orally in 2 human subjects, partial inhibition of intestinal motility stimulated by 5-HTP was also encountered (fig. 4).

In 2 subjects serotonin and 5-HTP in doses of 10 and 30 mg., respectively, were administered into the intestine proximal to the recording end of the intestinal tube. No effect on the intestine was noted.

It was reported previously that urinary excretion of 5-HIAA in 9 normal subjects was in the range of 2.1 to 5.2 mg. per day. Also, numerous specimens in patients whose diagnoses were in doubt fell within a range of 2.0 to 9.0 mg.[11] Blood platelet serotonin in 9 normal subjects ranged from 0.17 to 0.5 μg. per mg. of platelet protein.[12] In patients with unrelated diseases occasional values up to 0.9 μg. per mg. of platelet protein were found.

The results of the determination of platelet serotonin and urinary 5-HIAA in this study are summarized in table 1. Striking among these find-

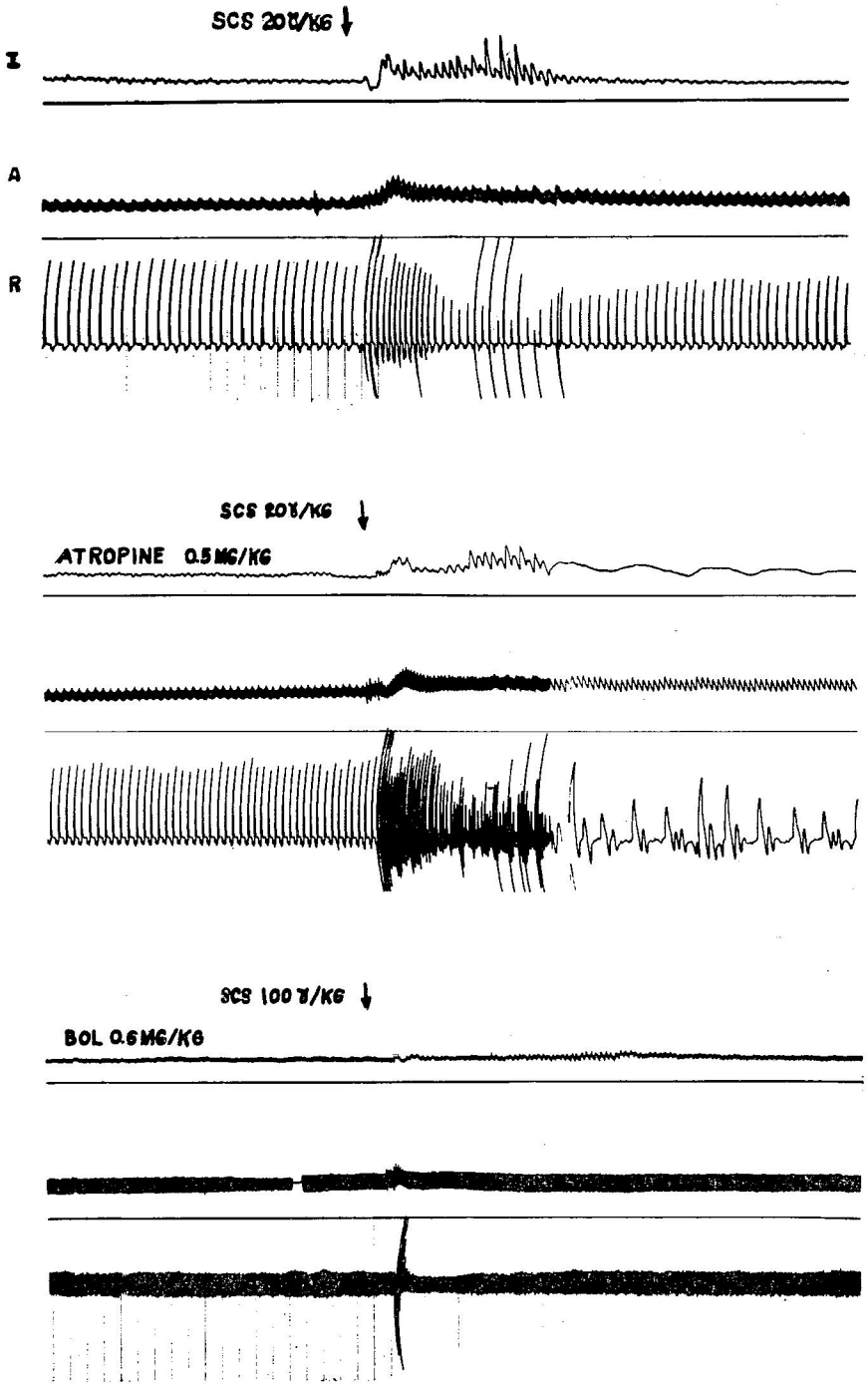

FIG. 3. Jejunal motility (*tracing I*), arterial pressure (*tracing A*), and respiration (*tracing R*) in the dog. *Brom*-lysergic acid diethylamide inhibited the responses to serotonin, but atropine sulfate was ineffective.

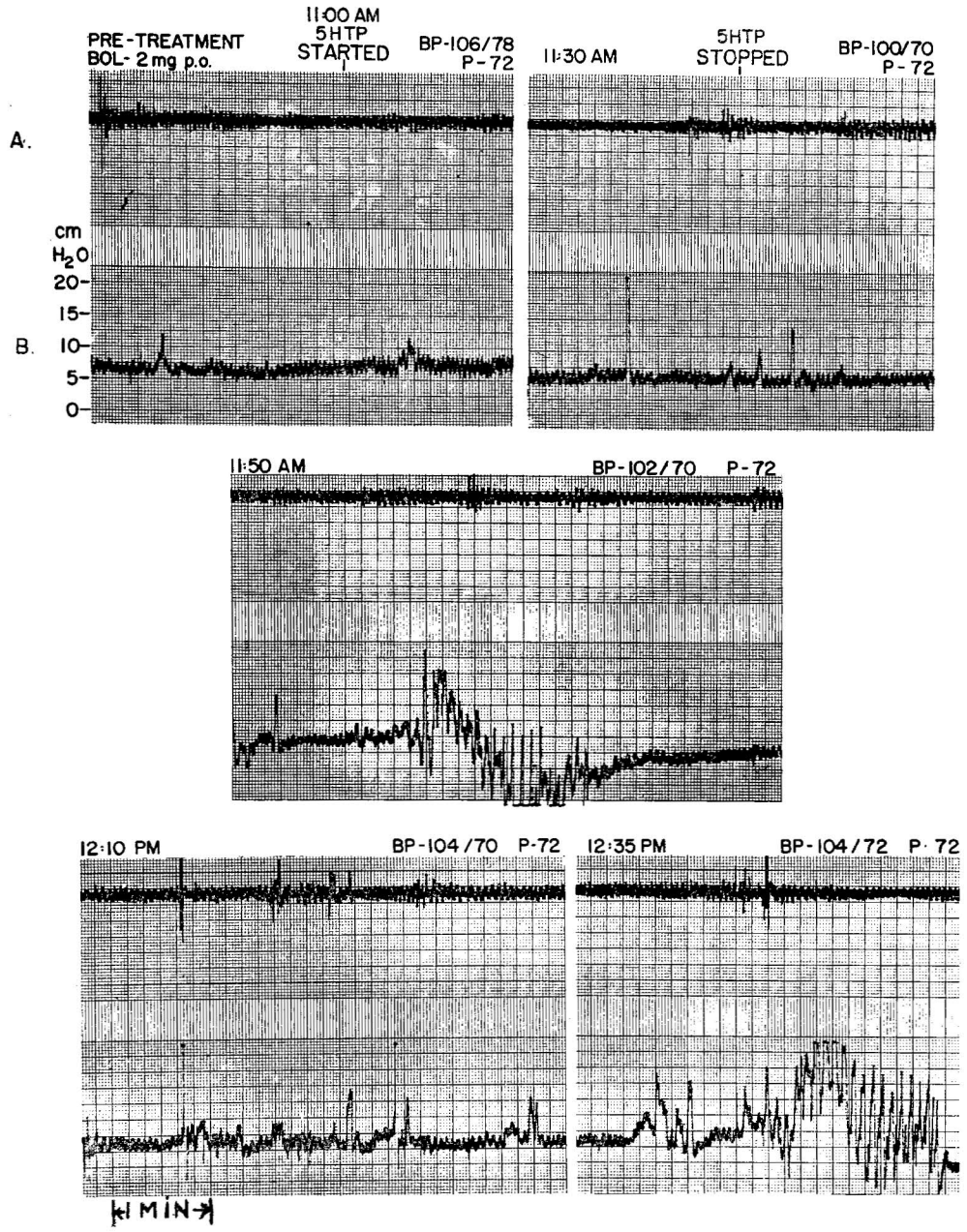

FIG. 4. Respiration (*tracing A*) and jejunal motility (*tracing B*) in a control subject. Pretreatment with *brom*-lysergic acid diethylamide partially inhibited motility stimulated by 5-hydroxytryptophan (5-HTP).

ings was the lack of any significant measurable amounts of blood serotonin and urinary 5-HIAA in the patient whose intestinal tract had been resected. Blood serotonin values on two occasions were less than 0.05 and 0.03 μg. per mg. of platelet protein. Four 24-hour 5-HIAA values in this patient were 0.35, 0.7, 1.06, and 0.95 mg. The 2 patients with nontropical sprue and the patient with Whipple's intestinal lipodystrophy had some blood serotonin and urinary 5-HIAA

TABLE 1

Blood serotonin and urinary 5-hydroxyindoleacetic acid (5-HIAA) levels in patients with various gastrointestinal disorders

Diagnosis and Patient	Blood Serotonin	Urinary 5-HIAA
	μg./mg. of platelet protein	*mg./24 hr.*
Nontropical sprue		
T. D.	1.25, 0.61, 1.57, 0.47, 1, 1.35	10.4, 10.4, 5.8, 10, 8.3, 5.8, 8.3, 5.8, 9.5, 9.5, 5.8, 5.8, 7.7, 6.5, 10.6, 10, 9.5, 11.9, 9.5, 9.5, 11.2
R. A.	0.40, 1.26, 0.28, 0.30	7.1, 7.7, 9.8, 7.1, 8.8, 9.2, 11.6, 10.9, 12.4
Intestinal lipodystrophy		
T. E.	0.49, 0.71	10, 9.5, 12.4, 10.6, 9.5, 8.4
Resection of large and small intestine		
F. W.	<0.05–<0.03	0.35, 0.7, 1.06, 0.95
Ulcerative colitis		
F. B.	0.16, 0.31	2.5, 1.8, 1.5, 2.6
C. R.		1.5, 4.3
Regional enteritis		
J. R.		1.6, 1.5, 1.8, 1.4
N. W.		3.4, 3.8
Normal range	0.17–0.7	2–9

values which appeared to be borderline elevations. The patients with regional enteritis and ulcerative colitis had normal values for blood serotonin (measured only in patient F. B.) and relatively low excretion of its urinary metabolite.

When urinary extracts from patient R. A. were subjected to paper chromatography for assay of 5-HIAA, large amounts of another indole compound were discovered. By methods described by Weissbach *et. al.*[13] this substance was shown to be indole-3-acetic acid. The daily excretion of indole-3-acetic acid by this patient was on the order of 200 mg. per day, whereas the usual range in a variety of other conditions was 3 to 20 mg. per day.[14]

Discussion

The results of this study indicate that intestinal motility is stimulated by a dose of 5-HTP which does not alter respiration, blood pressure, pulse rate, or mental status. Thus, the intestinal tract is extremely sensitive to the effects of serotonin. It is not unreasonable to conjecture that the largest store of serotonin in the body performs its function in the organ system where this store is also formed. Masson[14] many years ago demonstrated that enterochromaffin cells are in close proximity to the nerve endings of the enteric plexus. If the enterochromaffin cells normally influence intestinal motility, it is probable that they do so by releasing a humoral agent which acts on the neural elements within the intestinal wall.

When serotonin is administered by continuous infusion, the gut soon loses its ability to respond despite continued administration of the drug. In contradistinction to this, the administration of 5-HTP provides a sustained stimulus to intestinal motility. It is likely that this is attributable to the effect of intracellular serotonin, which apparently is released at a rate that permits a sustained elevated turnover rate of serotonin locally at the neural effector sites within the intestinal wall.

Bülbring and Lin[15] reported that the introduction of serotonin into the lumen of isolated loops of guinea pig's ileum stimulated peristalsis. They found that low concentrations of serotonin administered to the outside of the intestine never increased peristalsis and that high concentrations inside never abolished peristalsis. Their evidence suggested that serotonin may act *in vitro* by sensitizing the sensory receptors in the mucosa which trigger the peristaltic reflexes. However, in 2 subjects, when serotonin or 5-HTP was administered into the intestine no response was

evoked. Hendrix *et. al.*,[5] observed similar findings with serotonin.

It is of interest that atropine inhibits intestinal motility stimulated by 5-HTP in a dose up to 40 mg., but it did not modify the response of the bowel to serotonin creatinine sulfate in doses as little as 1 or 2 mg. As it has been generally accepted that 5-HTP exerts its action only after being converted to serotonin *in vivo*, this finding could only be explained by a difference in dosage or in the site of action of the two compounds.

In a previous report[4] it was demonstrated that lysergic acid diethylamide potentiates intestinal motility stimulated by serotonin despite the reported inhibitory properties of this substance on other actions of serotonin. BOL, which does not have the hallucinogenic properties of its analog, inhibits intestinal motility stimulated by either serotonin or 5-HTP.

The absence of significant amounts of serotonin and its metabolite in a patient whose intestinal tract had been resected supports the thesis that 5-hydroxyindoles in blood and urine represent mainly the contribution of the gastrointestinal tract. The futility of attempting to evaluate the status of brain serotonin by similar measurements is apparent. To date, significant changes in blood serotonin or urinary 5-HIAA appear to result from serotonin-producing tumors, alterations in gastrointestinal serotonin, administration of rauwolfia alkaloids,[12] or a dietary factor.[16] Further study is required to evaluate the significance of the elevations of 5-HIAA in the patients with nontropical sprue and Whipple's intestinal lipodystrophy and the relatively low values in patients with ulcerative colitis and regional enteritis. Similarly, the marked elevation of urinary indole-3-acetic acid in 1 patient with nontropical sprue suggests that further investigation of urinary indoles in intestinal disorders may be worth while.

Summary and Conclusions

1. 5-Hydroxytryptophan (5-HTP) is a potent stimulus to intestinal motility in man. This substance stimulates motility in a dose that does not alter respiration, blood pressure, pulse rate, or mental status.

2. 5-HTP provides a sustained stimulus to intestinal motility in contrast to the response following serotonin which is characterized by tachyphylaxis.

3. The intestinal tract makes a major contribution to the levels of blood serotonin and urinary 5-hydroxyindoleacetic acid (5-HIAA).

4. Patients with nontropical sprue and intestinal lipodystrophy had slight elevations in urinary 5-HIAA. One patient with nontropical sprue had a greatly elevated level of urinary indole-3-acetic acid. These findings indicate the advisability of further studies on indoles in patients with gastrointestinal disease.

Dr. Bernard J. Haverback
University of Southern California
School of Medicine
1200 N. State St.
Los Angeles, Calif.

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DISCUSSION OF PAPER PRESENTED BY DRS. HAVERBACK AND DAVIDSON

DR. THOMAS R. HENDRIX (Baltimore, Md.): Dr. Haverback's paper raises many interesting questions, but I will limit my discussion to only three. First: How does 5-hydroxytryptamine (5-HT) modify the motor function of the intestine? Two possible mechanisms have been suggested by experiments with isolated intestine. One suggestion, supported by studies by Gaddum, Rocha e Silva, and others, is that 5-HT stimulates intestinal smooth muscle by acting through "tryptamine receptors" on the postganglionic parasympathetic neurones. The other mechanism, suggested by Paintal and further developed in the elegant experiments of Bülbring and Lin, is that 5-HT increases intestinal tone and motility by lowering the threshold of the stretch receptors of the intestine, which in turn increases the sensitivity of the peristaltic reflex.

I wonder if Dr. Haverback would comment on whether or not his observations in man can be explained by either of these postulations based on studies *in vitro.*

The second question deals with the effect of atropine on the action of 5-HT. Reports of studies on isolated intestine have disagreed; some find that anticholinergic drugs block 5-HT action, whereas others can find no effect. The disagreement has now been extended to observations *in vivo* as well, for in our studies in man we found that anticholinergic agents clearly inhibited the action of exogenous 5-HT on the gut. These divergent results may be attributed in part to use of different amounts of 5-HT and anticholinergic drugs and in part to different recording systems. But are the conclusions presented justified when the records presented show atropine was given after 5-hydroxytryptophan (5-HTP) but before 5-HT?

Finally, I would like to ask Dr. Haverback if he believes the altered intestinal motility observed in nontropical sprue is related in any way to abnormal 5-HT metabolism. The disturbed intestinal motility in sprue has been considered secondary to malabsorption, but maybe studies of serotonin action may lead to an understanding of the specific defect.

DR. DHODANAND KOWLESSAR (New York, N. Y.): I would like to report confirmation of one phase of Dr. Haverback's studies; namely, the elevation of 5-hydroxyindoleacetic acid (5-HIAA) in nontropical sprue. Like Dr. Hendrix, we became interested in the relationship between the borborygmi, abdominal distention, diarrhea, and the metabolism of serotonin. We therefore began a systematic study of the urinary excretion of 5-HIAA in a variety of steatorrheal states as adjudged by increased excretion of I^{131} triolein.

To our surprise we found normal levels of 5-HIAA in all diarrheal states except nontropical sprue and the carcinoid syndrome. In the former group we found levels as high as 17 and 18 mg. of 5 HIAA, *i.e.,* two to three times our normal levels.

We followed about 7 patients with active nontropical sprue and elevated levels of 5-HIAA prior to treatment with the gluten-free diet and noted that when they were in biochemical remission as adjudged by normal I^{131} triolein, normal calcium, total protein, albumin, etc., the levels of 5-HIAA had also returned to normal.

I should like to call attention to an article in the *Proceedings of the Society for Experimental*

Biology and Medicine (February, p. 152, 1958), in which a group of workers in India were able to demonstrate increased levels of 5-HIAA in patients with schizophrenia. Whether the abnormal mentation that one frequently encounters in patients with nontropical sprue is intimately tied up with abnormal tryptophan metabolism, we do not know.

CONCLUDING DISCUSSION BY DR. HAVERBACK

DR. BERNARD J. HAVERBACK (Los Angeles, Calif.): I should like to amplify one point that Dr. Hendrix mentioned.

Bülbring has found that the introduction of serotonin into the lumen of guinea pig ileum *in vitro* stimulated peristalsis but not when administered to the outside of the intestine. The evidence suggested that serotonin may act *in vitro* by sensitizing the sensory receptors in the mucosa which trigger the peristaltic reflex.

Considering the anatomic proximity of the enterochromaffin cells to the terminal fibers of the submucus plexus, it is likely that serotonin regulates intestinal motility by acting on neural elements within the intestinal wall.