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# Celiac disease : the disease, its pathology, and the history and use of the peroral biopsy tube in the diagnosis

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# CELIAC DISEASE: THE DISEASE, ITS PATHOLOGY, AND THE HISTORY AND USE OF THE PERORAL BIOPSY TUBE IN ITS DIAGNOSIS

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# INTRODUCTION

"There is a kind of chronic indigestion which is met with in persons of all ages, yet is especially apt to affect children between one and five years old. Signs of the disease are yielded by the faeces; being loose, not formed, but not watery; more bulky than the food taken would seem to account for; pale in colour, as if devoid of bile; yeasty, frothy, an appearance probably due to fermentation; stinking, stench often very great, the food having undergone putrefaction rather than concoction." (20)

Thus began the classic account by Doctor Samuel Gee of a specific type of diarrhea especially found in children. The true etiology of the idiopathic type of steatorrhea is as obscure today as it was in 1888, when Gee wrote his article. Although quite brief, Gee's picture of the disease was so perfect that there has been little to add to his clinical account. The symptoms he described were, first and foremost, the passage of loose, pale, bulky and offensive stools on a normal diet. The abnormal stools may be consistent in their number and degree of abnormality, or they may appear in exacerbations associated with respiratory or other infections. In more severe cases, physical signs of malnutrition appear which are in no sense specific to this disorder but represent indications of chronic

starvation. They include (1) retardation of growth, (2) wasting, particularly of the extremities, buttocks and trunk, (3) abdominal distention, (4) associated nutritional deficiencies, commonly including iron deficiency with a characteristic macrocytic hypochromic anemia.

* * * * *

Since this pattern of clinical findings may result from a variety of disturbances, it is termed the celiac syndrome. Nelson's Textbook of Pediatrics (36) divides this into the following disease entities: (1) true or idiopathic celiac disease, also called the malabsorption syndrome; (2) celiac syndrome resulting from mechanical obstruction of digestion and/or absorption; (3) pancreatic insufficiency (cystic fibrosis of the pancreas); (4) severe dietary deficiency (starvation, kwashiorkor); (5) gastrointestinal allergy; (6) chronic intestinal infection (bacterial, parasitic, viral).

Certain laboratory findings are helpful in the diagnosis of the celiac syndrome. The fat content of the stool is high. The normal child excretes fat in a quantity up to 20% of the dried volume of the stool. In patients with celiac syndrome, the fat content is likely to range from 30 to 50%. An average output of fecal fat of less than 5 grams a day on a daily fat intake of more than 50 grams may be taken

to exclude the diagnosis of celiac syndrome. In this condition the fecal fat output exceeds 5 grams a day and commonly averages more than 10 grams (33).

The absorption of vitamin A is also altered. Measurement of the amount of vitamin A in the blood plasma following the administration of a test dose may be used to indicate the efficiency of the intestine in absorbing fat. A test dose (0.1 cc per pound of body weight of a fat-soluble vitamin A concentrate containing 60,000 I.U. per gram) is given orally, and blood samples are taken before the test dose and at 3, 5, 7 and 12 hours after. Normally, the peak of the post-absorptive levels occurs in 3 to 5 hours and is from 5 to 10 times that of the fasting level. In all varieties of the celiac syndrome, the curve of vitamin A levels is lowered, and in severe cases no rise occurs.

Oral glucose tolerance tests show a low rise of less than 30 mg% in patients with the celiac syndrome except in those due to fibrocystic disease. The neutral fat level of the blood is reduced, and the total cholesterol level is likely to be in the lower range of normal (100-130 mg%).

Radiography may also be of some help. Many patients with the celiac syndrome show gross clumping of barium sulfate when a simple suspension of this opaque medium in water is used. This appearance is, however, not uncommon in

normal children, although usually to a lesser degree.

Examination of the small bowel contents is another good diagnostic procedure. Samples of the contents of the upper small intestine are withdrawn by intubation. The contents are examined for pancreatic enzymes, bile salts, and organisms. A stimulus to pancreatic secretion, such as secretin may be given. Pancreatic enzymes should be studied individually, since simple enzyme defects may occur.

Cooke, Peeny, and Hawkins (11) believed that persistently normal blood levels were strong evidence against the diagnosis of idiopathic steatorrhea. Two-thirds of their patients had macrocytosis when their blood was first examined, and the remainder developed an increased mean cell volume at some time while under observation. Other clinicians have also found similar findings. A normochromic macrocytic anemia or a hypochromic anemia in which there was considerable depth of staining is suggestive of steatorrhea and was often the first clue to the diagnosis.

More note has been taken of iron deficiency because iron absorption is defective and many patients respond poorly to oral iron therapy. Parenteral iron will, however, restore the picture when macrocytosis is likely to become evident (25). Though defective absorption seems an adequate explanation, studies with oral iron absorption tests suggest that there may be an additional factor, failure to utilize

the iron, for by calculation from the iron absorption curves sufficient iron should be absorbed to allow adequate hematological response.

There is a group of infants who present the picture of celiac syndrome in which no clear-cut cause can be determined and the diagnosis is made largely by the process of exclusion. This group is considered to be suffering from idiopathic celiac disease. Characteristically, these infants do not show the typical signs until the end of their first year of life and are involved most severely during their second year. The first indication of abnormality usually follows an acute parenteral infection, when the diarrhea associated with the infection fails to clear. In mild cases, the characteristic persistence of digestive upset after each infection may be the only indication of abnormality. In others, the diarrhea persists at all times.

The defect in these children is one of absorption rather than digestion. Fat and carbohydrate absorption are particularly involved, while protein absorption is little affected. The basic defect is obscure. Increasing interest is being given to the question of a family predisposition since Ebbs (15) in 1950 suggested that the incidence among relatives was too high to be a matter of chance. This suggests the possibility of a genetic origin. Ebbs theorized that the incidence of diabetes mellitus in families of

children with celiac disease was sufficiently high to link the condition with the diabetic trait.

During the last few decades, clinical observations have emphasized the similarity of the gastrointestinal dysfunction in children with idiopathic steatorrhea to that seen in non-tropical sprue, and it is now recognized that the numerous adult patients with non-tropical sprue represent the continuing manifestations of the childhood disorder (19).

* * * * * *

Since the development during the past few years of the intestinal biopsy tube, interest has returned to the pathological findings found in the intestines of patients with idiopathic steatorrhea and non-tropical sprue. This search for both the etiologic basis of the disease and the application of the pathologic findings as a diagnostic tool for definitive diagnosis of the syndrome was necessary; as previously mentioned, present diagnosis depends on the exclusion of the other causes of the celiac syndrome.

Interest in the pathology of this disease dates back to the classical work of Gee who stated in his monograph: "Whether atrophy of the glandular crypts of the intestine be ever or always present, I cannot tell" (20). The small intestine findings are logically the chief points of interest. A review of the literature reveals considerable

divergence of opinion as to the nature of pathologic changes and even as to the presence or absence of such changes.

Thin (51) and Weathered (54) in 1890 found atrophic changes in the small intestine, but the latter viewed these with some skepticism and thought them due to post-mortem changes. Faber (16) in 1904, and Justi (28) in 1913 described ulcerations with secondary widespread destruction of the villi. Justi reported that the villus structure was short and swollen, that the free one-half of the stoma contained many chronic inflammatory cells, and that the core of the villus was filled with very fibrillar cellular connective tissue. Beneke (4) in 1910, however, did not agree with these findings, and felt that marked inflammatory changes were not present. Faber, Van der Scheer, Thaysen and Mackie found no such alterations, although the latter spoke of a withering of the villus. Manson-Bahr (29) in 1914 considered the villi atrophic from proliferation of fibrous connective tissue, and thought similar changes were present in the submucosa. He also reported that the mucosa was diffusely infiltrated with round cells.

In 1922, Manson-Bahr (30) thoroughly studied eight cases of adult sprue and decided the earliest changes were in the villi where there was infiltration with small round cells and a degeneration of the epithelial covering. Some congestion was also noted, but signs of active inflammation

or infiltration with polymorphonuclear leukocytes were absent. Essentially, he considered the process a degenerative and non-inflammatory one, the end stage being a shrunken and acellular villus. Most of the changes were in the jejunum and, although occasionally ulcerations without inflammatory reactions were seen, no scars were observed. Eventually, although apparently not early, the glands of Lieberkuhn underwent degenerative changes. In half the cases, he noted an atrophic thinning of the gut wall.

Thaysen's monograph (49), published in 1932, dealt extensively with sprue. His observations led him to the conclusion that there were no morbid histologic changes in the intestine in either non-tropical sprue or its childhood counterpart, idiopathic steatorrhea. He dismissed all the strong evidence to the contrary advanced by the previous workers of inflammation and ulceration of the intestinal mucosa, because they had not constantly been seen or were autolytic. Elsewhere, however, he said: "It must be correct to interpret these intestinal changes as being of secondary nature, either produced by relatively low virulent micro-organisms that have nothing to do with the aetiology, or resulting from irritation of the mucous membrane owing to abnormal acidity of the intestinal content, or as a result of both agents" (49).

Thaysen mentions only four cases of idiopathic steatorrhea in which microscopy of the intestine had been carried out. His review included 14 autopsied cases of tropical sprue and 34 cases of non-tropical sprue from the literature; but in only 6 of the latter were there autopsy findings to be analyzed, and in one of these there was no histological description of the intestines. As a result of the acceptance of Thaysen's belief that the jejunal mucus was normal in idiopathic steatorrhea and non-tropical sprue and that any abnormality found should be attributed to post-mortem changes, for twenty years those interested in this field ignored the possibility of a mechanical hinderance to absorption and sought, without success, for causes ranging from vitamin deficiency to adrenal dysfunction. It was not until 1954 that Paulley (34) reviewed Thaysen's monograph and disproved it on the basis of a rather one-sided review of the literature and insufficient microscopic findings to substantiate his beliefs.

Paulley believed that the only way of testing the jejunitis hypothesis and avoid post-mortem autolytic changes was to obtain biopsy material. He obtained laparotomy biopsies from four patients diagnosed as having non-tropical sprue of whom one during childhood presented good symptomatology for a diagnosis of idiopathic steatorrhea. He found in all four cases inflammatory changes in the upper

small bowel. The microscopy findings of the mucosa were striking. The villi were approximately double the width of normal control samples, and other differences were present in the shape and cellularity of the villi, the presence of large numbers of inflammatory cells in the mucosa, and extending into the submucosa, fibrosis and dilitation of the lymphatics. There was also some histologic evidence of edema, and the delicate appearance of the normal jejunal mucosa as seen in control material was missing. Goblet cells appeared more prominent in the steatorrhea cases than in the controls.

In reference to previous workers' findings, Paulley stated, "So seldom have pathologists been confronted with biopsies of human jejunum that it is doubtful if the normal appearances are much known. The shape, stroma, and cellularity of the normal as opposed to the abnormal villus is likely to be difficult to interpret and to call for experience at present lacking."

For these reasons he believed he could understand why these mucosal changes had been found only inconsistently in the preceding years with the occasional material, and that being usually severely damaged by autolysis. To Paulley it seemed that the inconsistency of the mucosal findings in the past may have been due to ignorance of the normal appearances, differences in the stage of the disease, post-

mortem or even ante-mortem changes making reliable interpretation impossible, or to absence of biopsy material.

It must be concluded that the mucosa of the gastrointestinal tract in patients with celiac disease appears to be unusually susceptible to post-mortem autolytic changes. For this reason, the interpretation of various changes seen in the small bowel at autopsy is difficult. Consequently, the more recent utilization of biopsy techmiques in the study of this disease has enabled the pathologist to interpret changes which are not influenced by autolysis. The oral technique of obtaining biopsy material represents an important advance in the study of small intestinal pathology. The factors of agonal changes and post-mortem autolysis are eliminated, thus permitting a more exact interpretation of pathologic changes in the small bowel.

* * * * * *

During the past ten years, many articles have appeared describing the use of small-bowel biopsy tubes, which for the first time enable the clinician to obtain tissue from the small bowel with a minimum of discomfort to the patient. Since these are peroral techniques which do not require anesthesia, manipulation or clamping of the organs, and since fixation is accomplished seconds after the tissue is removed, excellent histologic preparations can be made. The use of

peroral suction biopsy tubes to obtain specimens of gastrointestinal mucosa has become widespread since Wood and his associates (55) and Tomenius (52) introduced the principle with their gastric biopsy tubes. In recent years the method has been extended to obtain mucosal specimens from the small bowel and colon.

The original Wood's biopsy tube (Figure 1) perfected in 1949 consisted of an outer casing of Bowden wire in a closely wound spiral (2½ mm. outer diameter) of 28-gauge stainless steel inserted in a close-fitting plastic tube. The proximal end was fitted to a vacuum-tight gland through which passed the inner operating wire of 22-gauge stainless steel with handle. A lateral tube allowed connection to a hand vacuum pump through a rubber tube. The distal end was fitted to a stainless-steel cylinder lapped to fit the knife. The knife was screwed onto the end of the operating wire which retracted it, and down past a 3/32-inch-diameter countersunk hole drilled in the cylinder wall. The cylinder end was fitted with a removable screw which allowed the knife to be pushed out and unscrewed for obtaining the specimen, cleaning and sharpening. The tube was about 100 cm. long. Specimens usually included the muscularis mucosa. No complications were noted from use of the tube. Wood subsequently modified the tube by increasing the inside diameter from the 2½ mm. to 3 mm. and was able to get a higher

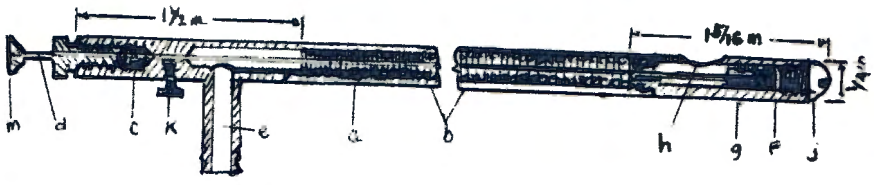



FIGURE 1

WOOD'S FLEXIBLE GASTRIC BIOPSY TUBE: a) Bowden wire casing; b) plastic tube; c)vacuum-tight gland; d) operating wire; e) lateral exhaust tube; f) steel cylinder; g) cylinder knife; h) hole; j) terminal screw; k) fexation screw; m) handle; Lancet, 1:19, 1949

percentage of biopsies.

In 1956, Shiner (41) contended that a modified Wood's tube might be passed beyond the pylorus to sample duodenal or even jejunal mucosa, and that this technique might be of diagnostic value in such conditions as steatorrhea. Her duodenal biopsy tube (Figure 2) was a flexible tube, 128 cm. long and 4.8 mm. in diameter, coated with plastic material to make it smooth and air tight. Its head piece was a small tube, 3.9 mm. in external diameter, at whose center was the circular biopsy aperture 3.1 mm. in diameter. In this small tube the knife cylinder was accommodated which was screwed to the wire running along the whole length of the instrument and ending in a small holder at the tail piece. The small tube of the head piece was imbedded in a solid, weighty, elliptical, nickle ball 12.3 mm. in diameter to give smooth easy forward propulsion by gastric and duodenal peristalsis. The tail piece contained the other end of the wire and the latter exhaust tube, to which a syringe or a pressure manometer could be attached to apply negative pressure. Fluoroscopy was used for guiding the course of the tube. No complications were noted in 19 duodenal biopsy attempts, 12 being successful.

Shiner (42) modified this tube for jejunal biopsy (Figure 3) by increasing the length to 161.5 cm. and the diameter to 5 mm. The head piece was changed to 2.5 cm. in length

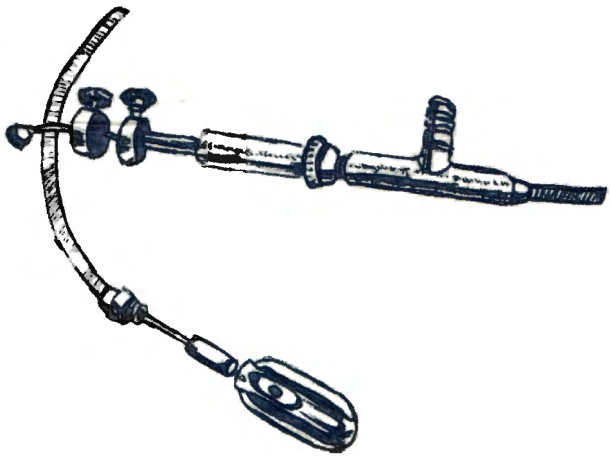


FIGURE 2

SHINER. DUODENAL BIOPSY TUBE: headpiece and tailpiece Lancet 1:17,1956

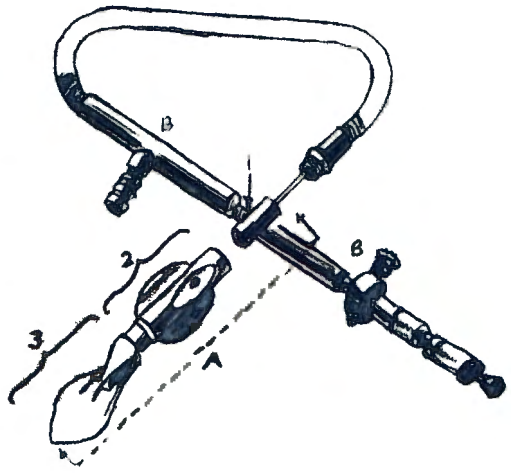
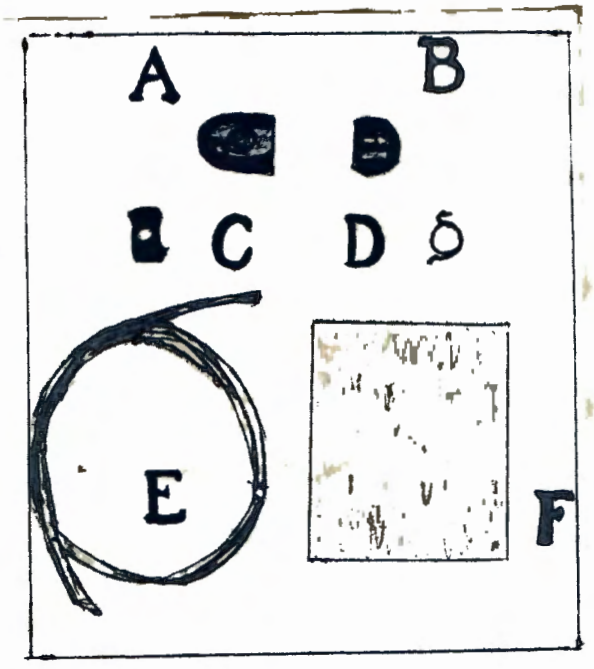



FIGURE 3

SHINER JEJUNAL BIOPSY TUBE: A, headpiece: (1) knife cylinder (2) biopsy head: (3) Balloon: B, tailpiece, with lateral exhaust. Lancet, 1:85, 1956.

and had a diameter of only 1.3 cm. at its widest point. The diameter of the biopsy hole was 3.5 mm. The knife cylinder was 9 mm. long and ridged at its distal end. An air hole was situated in one of its ridges extending down the side of the knife cylinder, thus allowing air to reach the rubber balloon at the distal end of the head piece. The tail piece was essentially the same as in Shiner's previous tube except that it was longer and allowed more play in the movement of the wire, which had to travel a greater length within the flexible tube. When the tube reached the first part of the duodenum, the balloon was inflated and used to guide the head piece along into the jejunum. Shiner reported the balloon as essential; otherwise she found it impossible to travel through the second and third parts of the duodenum.

In 1957, further modifications by Crosby (13) resulted in an intestinal biopsy capsule (Figure 4) which is still in use. The instrument essentially consists of a cylindrical stainless-steel capsule 20 mm. in length and approximately 11 mm. in diameter, divided in circular cross section by a rubber diaphragm. In the proximal section, a knife blade sweeps the wall of the capsule through an arc of 90 degrees. The distal section is empty and serves as a pneumonic cell. The rotary movement of the knife block is activated by a




Disassembled

Assembled

FIGURE 4

CROSBY BIOPSY CAPSULE: chambers A and B; C the knife block; D the wire spring that activates the knife block; E the polyethylene tubing; F a latex rubber dam.

spring coiled about the axis. The block with its knife is held in a cocked position by a key on the wall of the proximal section which fits into a slot on the outer surface of the block. The knife is sprung by moving the block in a linear direction until the key escapes from the slot and the block is then free to pivot. The knife is moved in a linear direction by a bulging of the rubber diaphragm between the chambers.

The port of entry of the biopsy sample is 5 mm. in diameter. This accommodates an adequate piece of mucosa without danger of dimpling and nipping the muscularis. There is a second 2-mm. perforation at the base of the axial shaft into which is fixed the end of a polyethylene tube.

To activate the knife suction is applied to the proximal chamber through the polyethylene tube. Intestinal mucosa adjacent to the port is sucked into it, thereby occluding the port. Suction continues and the pressure in the proximal section becomes less than that in the distal section. The rubber diaphragm bulges in the proximal section pushing the knife block in a linear direction and thereby releasing the spring activated mechanism. The knife blade snaps shut, amputating the mucosa which has herniated into the proximal capsule. When the knife has been sprung, the block closes the port, preventing loss of the sample. Only single biopsy specimens are possible with each intubation.

Crosby found this tube to be both reliable and safe. It is technically uncomplicated and it imposes a minimum of hardship on the subject.

Also in 1957, Shiner again added minor modifications to her tube and published the results of a series of 45 biopsies (43). The tail piece of the modified apparatus was fitted with a solid rod fused to the long wire inside the flexible tube. A special holder ensheathed the solid rod so that its movements were accurate and easy. The lateral exhaust tube of the tail piece was attached to a pressure manometer graded in inches of Hg. Shiner used a pressure of 3 to 5 inches Hg. on the manometer and stated it dangerous to exceed a pressure of 10 inches Hg.

In 69 biopsy attempts, Shiner reported the following causes for failure:

1. Failure to introduce the head piece beyond the hypopharynx, 2 times.

2. Failure to pass beyond the pylorus. This presented the biggest problem with Shiner's tube and occurred 18 times.

3. Failure to obtain a full thickness of mucosal specimen, 6 times.

4. Autolysis of the specimens, 5 times. This apparently was due to the presence of large amounts of intestinal juices inside the biopsy tube combined with too long a

delay in withdrawing the tube.

5. Failure to obtain a specimen once the head piece had reached the desired level. This occurred in 12 patients and was thought to be caused by a leak in the instrument, failure to operate the knife blade satisfactorily, or the use of incorrect pressure.

Shiner stressed that in the 69 attempted biopsies, no complications occurred.

Of the 30 successful duodenal biopsies, one was from a 65-year-old woman previously diagnosed as having an idiopathic steatorrhea with a megaloblastic anemia. Included in the series of 9 successful jejunal biopsies was a 65-year-old male diagnosed as having a megaloblastic anemia of uncertain etiology, and a 65-year-old male diagnosed as having steatorrhea, megaloblastic anemia and subtotal gastrectomy for cancer of the stomach. Shiner noticed the striking similarity of these three specimens which showed almost total loss of the villi. Shiner suggested that the malabsorption syndrome of these patients was in part due to the tremendous loss of the villous surface epithelium. She further stated that jejunal biopsies could be of the utmost interest in the studies of the malabsorption syndromes of the small intestine.

Thus, with the development of working peroral biopsy tubes, Shiner became one of the first researchers to

substantiate Paulley's beliefs as to the pathology of the small intestine in steatorrhea.

Several months later in the same year, 1957, Butterfield and Perez-Santiago (8) reported their findings from operative jejunal biopsies from 4 patients with non-tropical sprue. Three patients were multiparous females at time of tubal ligation, and the fourth was a male patient biopsied at the time of laparotomy to rule out the possibility of Whipple's disease. All had shown steatorrhea with megaloblastic anemia previously, but were in remission at the time of biopsy.

All the biopsy specimens showed anatomic variations from the pattern normally seen in the jejunum. All the patients with sprue showed thickening and apparent shortening of the villi. The villi appeared to coalesce and to branch. They were often broad-ended and edematous. Goblet cells were increased. The columnar nuclei in some cases were fragmented and pyknotic. The mucosa was infiltrated by inflammatory cells, chiefly eosinophils. These changes could not be accounted for after careful review of the clinical records of the patients, and the observers were not inclined to attribute them to the surgical procedure.

Speculating as to the relationship between the anatomic findings and the factors interfering with intestinal absorption in these patients, the two researchers concluded

that the clubbing and coalescence of the villi must reduce the total absorbing area. Secondly, the changes in the cells themselves from healthy-looking cells to distorted and globular cells must interfere with absorption. They thought it possible that the movements of the villi were impaired, which would interfere with the pumping action of the villi. Finally, the congestion that they saw in the submucosal retion must impinge on the transport pathways away from the intestinal mucosal cells.

In July, 1958, Bolt, Pollard, and Standaert (5) published a report concerned with the use of the Shiner biopsy tube as an aid in establishing the diagnosis of small bowel disease. In their series of 34 patients, adequate biopsy specimens were obtained from 30 patients. The biopsies were accomplished easily and safely, and without any complications.

The characteristic findings of non-tropical sprue or idiopathic steatorrhea were noted in 7 patients; in all the presence of steatorrhea was proved on the basis of balance studies, as well as by the use of radioactive triolein $I^{131}$ methods. The biopsy specimens of these patients were all similar in appearance and distinctly different from the appearance of normal small bowels. Marked atrophy of the mucosa was present. In addition, there was marked cellular infiltration of the tunica propria, flattening of the epithelial surface of the villi, and obvious coalescence of

the villi. It was extremely difficult to obtain any sections showing the crypts at surface epithelium extending down to the submucosa. It was noted that these findings had been reported previously by others, and concluded that in their experience the changes described had been limited to the syndrome of non-tropical sprue or idiopathic steatorrhea. The investigators were impressed with the fact that they had not as yet encountered a normal appearing mucosa in the presence of non-tropical sprue; and likewise, had not encountered the histologic findings of non-tropical sprue either in normal controls or in patients with gastrointestinal disease other than non-tropical sprue or idiopathic steatorrhea.

In August of 1958, Himes and Adlersberg (26) reported 15 cases of intestinal findings, 11 studied at autopsy and 4 biopsied with the Shiner biopsy tube. These were 15 cases of idiopathic sprue in which the diagnosis was established by accepted clinical and laboratory criteria. None of these patients could be considered as having tropical sprue. Two additional patients without sprue were also studied by the biopsy method for comparison.

Of the post-mortem speciments the most consistent gross change observed was atrophy and thinning of the wall of the small bowel. This was seen in 7 cases. In the remaining 4 cases the small bowel was normal on gross examination. In 2 of these latter 4 cases, microscopic examination revealed

no striking abnormalities. Of the 7 cases exhibiting gross atrophy of the wall, 4 displayed significant microscopic abnormalities consisting of flattening and clubbing of the villi and increase in the mucosal cellularity. Instead of the normal elongated, finger-like villi normally seen, the structures were plump, short, and thick. In some areas the villi appeared flattened to the point of obliteration. There was considerable variation in the degree of cellularity, not only from patient to patient, but also within the same bowel. The cellular exudate consisted mainly of chronic inflammatory cells, often with many plasma cells and occasional eosinophils.

In the remaining cases, the interpretation of somewhat similar changes was difficult because of post-mortem autolysis.

All 4 of the biopsy specimens revealed pronounced changes in the mucosa. These changes were especially marked in the villous layer and consisted of various degrees of thickening of the villi, progressing from clubbing to complete fusion. The architectural alterations appeared to be more advanced near the periphery of the villi, whereas the deeper crypts and glands were more normal. The surface epithelium, instead of appearing as the usual tall columnar type was uniformly lower, being cuboidal in some areas. The nuclei of these cells were generally irregular in size and

position within the cells. There was a moderate increase in goblet cells in some instances. In all cases, the goblet cells were choked with mucus, a finding these workers considered compatible with the hypersecretion characteristic of sprue in roentgen studies. There was an increase in the cellular exudate of the lamina propria varying from slight to heavy. The cells were mainly lymphocytes and plasma cells with occasional eosinophils. There was some degree of swelling of the lamina propria in all cases.

One case of pancreatic steatorrhea and 1 case of pernicious anemia were biopsied, and the mucosal patterns were essentially normal in both instances.

Himes and Adlersberg concluded that significant pathologic changes are to be found in the small intestine of patients suffering from idiopathic steatorrhea or non-tropical sprue. They thought it possible that the morphologic changes in the bowel mucosa and the absorptive defect of sprue are related to each other as cause and effect.

They surmised that the earliest changes in the small intestine occur at the tips of the villi and consist of thickening and swelling (clubbing). Further progression of the process results in fusion of the villous tips, with complete obliteration of the normal pattern. This change results in an almost completely flat surface with marked reduction of the area available for absorption from the lumen of

the small bowel. The deeper structures show less alteration. These observations suggested to the investigators that the factors responsible for the changes, whatever they are, exert their influence primarily at the periphery of the villi.

In September of 1958, Crosby and several other investigators (45) published the results of 63 attempts to obtain biopsy material from 38 patients using the Crosby biopsy tube previously described. During the period of this instrument's development, failure to obtain a specimen occured in one-third of the attempts; however, these authors stated that at the time of publication their failure rate had been reduced to only 10%. Their reasons for failure were listed as:

1. Failure due to difficulty in swallowing the instrument, or in securing its passage into the desired level.

2. Failure of the instrument to "fire", which was almost always due to occlusion of the tubing by food particles, thick mucus, kinking of the tubing, or jamming of the moving parts.

3. Failure due to occlusion of the port by mucus or food when everything else went as it should, but no specimen was found when the capsule was opened.

Test firing the instrument before it is used and filling the capsule with air just before suction is applied

seem to be important in reducing the number of failures.

In their initial attempts with the Crosby tube, 63 attempts to obtain biopsy specimens were made on 38 persons with 22 failures. Thirty-three of the 41 specimens came from the jejunum, 2 from the ileum, 1 from the stomach, and 5 unintended came from the esophagus when the operator thought the tube to be in the stomach. No complications were experienced, even when as many as 3 successful biopsies were achieved from 1 patient. The observers noted that muscular mucosa was only infrequently present on the specimens as opposed to results with other earlier biopsy tubes.

The investigators assumed that the healing of the mucosal defect caused by the biopsy instrument took place by contraction of the muscularis mucosa which closed the defect and allowed healing to take place from the adjacent mucosa. As intestinal mucosa is one of the most rapidly regenerating tissues in the body, ultimate covering of the defect should take place within a matter of days.

The investigators were impressed that infection, hemorrhage or perforation had not been observed in any of their patients.

In discussing the practicality and usefulness of the Crosby biopsy tube, these workers thought that if due precautions were observed, the histologic sections obtained

would be excellent and would show no post-mortem changes when the preparations were studied under light microscopy. The finest detail of the mucosa could be discerned. They were also impressed with the uniformity of the findings in their normal subjects, but felt much greater experience would be needed to define the limits of normal and to elucidate the significance of borderline cases.

In contradistinction, the 8 successful jejunal specimens from the 4 patients previously diagnosed as having the malabsorption syndrome showed definite changes from the normal. There was blunting and fusion of the villi, variation in the size of the villi, and an increase in the cellularity of the lamina propria. These changes affected the individual villus to varying degrees, but the summation of the findings were thought to be such as to be diagnostic.

The newest member of the intestinal biopsy tubes, the multipurpose suction biopsy tube (Figure 5) was developed in 1959 by Brandborg, Rubin, and Quinton (7). These men felt that all the other biopsy instruments had limitations due to their large size, rigidity, or other mechanical defect. They felt the need for a safe and simple means of biopsying diffuse disease in any part of the gastrointestinal tract. They believed that their tube, although far from perfect, was a better one. Its flexibility and small diameter,

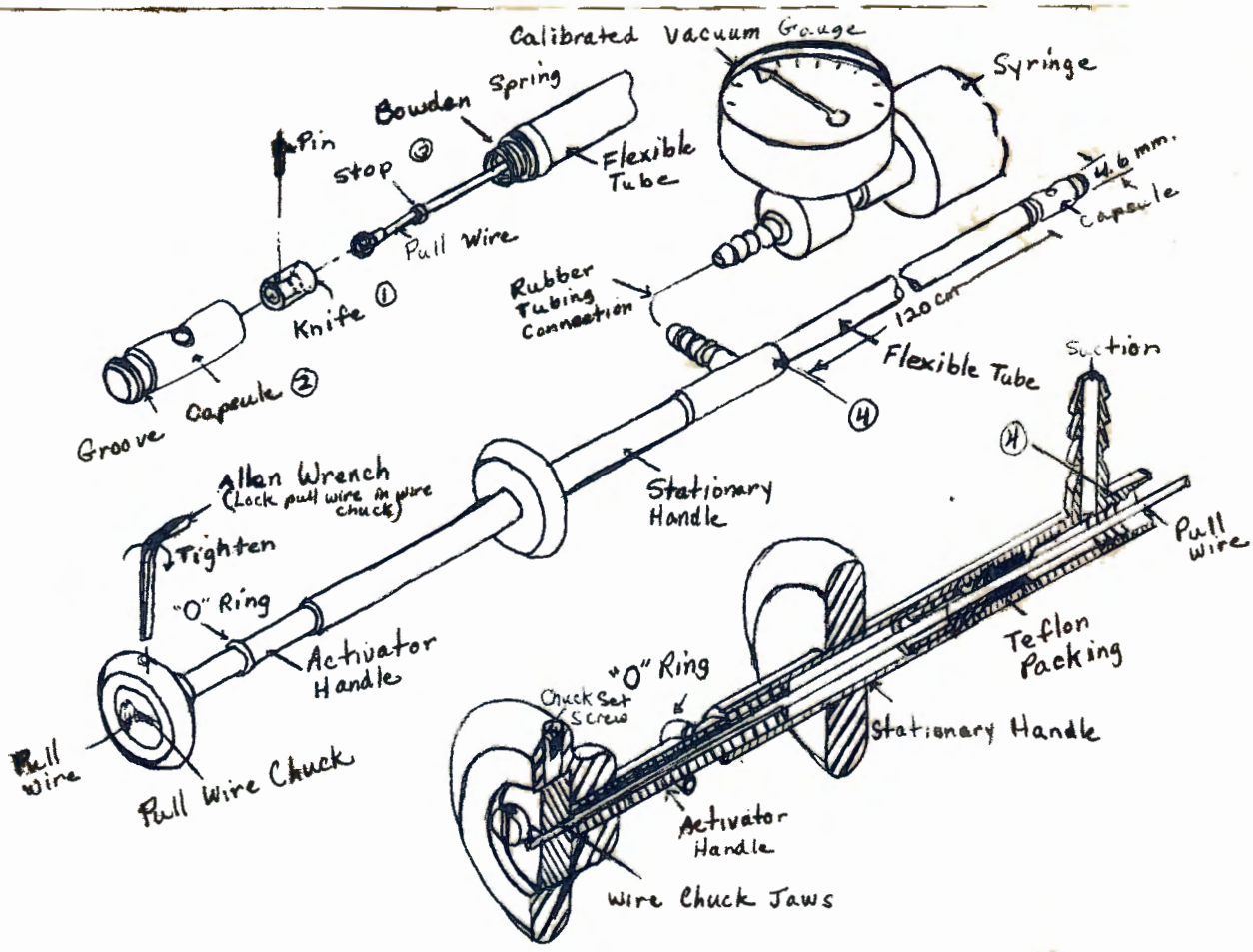



FIGURE 5

MULTIPURPOSE SUCTION BIOPSY TUBE:
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4.6 mm., enabled them to use it with equal facility in infants, children, and adults. Clean excision of multiple small biopsy specimens during a single intubation has been furthered by the present design which incorporates a precise, protected cutting mechanism, a leak-proof system, and a suction syringe attached to an aneroid manometer.

These investigators reported no occurrence of overt hemorrhage in over 700 biopsies with the new multipurpose biopsy tube, whereas they reported a 1% incidence in over 300 patients biopsied with other instruments. Despite this reduction in morbidity, they emphasized that occasional bleeding is a calculated risk which is acceptable because of the value of these procedures to the patient. With use of the proper technique, they reported, perforation is highly improbable; it has never occurred in over 1000 suction biopsies, including those taken with the earlier tubes.

In 70% of their biopsies done on adults, two specimens were obtained with one intubation. They also reported successfully taking three specimens on occasion, but in order to avoid traumatizing the specimens in the small capsule, they did not attempt more.

These workers were able to intubate most children over 6 years of age without restraints or sedation, provided an effort was made to establish rapport with the child. In

younger children, they immobilized the child by wrapping him in a sheet. Sedation in these younger children often produced irrational excitement unless given in large doses.

They reported an 82% success of biopsy attempts from all levels of the gastrointestinal tract at the time of their initial attempts, and a 90% success at the time of publication of their report. They reported 55 unsuccessful attempts during the first intubation, but in most of these patients a successful biopsy was achieved after a second intubation was attempted. Their reasons for failure were listed as:

1. Failure to biopsy esophagus through an esophagoscope, 4 cases.

2. Failure due to secretions or food present in the stomach, 16 cases.

3. Failure due to aluminum hydroxide present in the small bowel, 1 case.

4. Failure due to biopsy from gastric antrum instead of duodenum, 13 cases.

5. Failure due to inability to pass the pylorus, 4 cases.

6. Failure due to cystic fibrosis of the pancreas, 2 cases.

7. Failure due to biopsy of the rubber bag, 2 cases.

8. Failure of colon biopsy due to presence of stool, 10 cases.

9. Failure due to tube malfunction, 3 cases.

The 29 combined failures due to food or secretions in the stomach and from biopsy of the gastric antrum instead of the duodenum occurred in infants and small children, and the latter group represented failures of small bowel intubation. These latter failures were partially ascribed to stiffness of the earliest tubes and partially to the difficulty in intubating an uncooperative child. Tube malfunction occurred solely with the prototypes of the present tube.

Their more than 700 biopsy specimens taken with the multipurpose biopsy tube were in patients ranging in age from 3 months to 85 years. There had been no overt hemorrhage or other complications from these biopsies.

The researchers felt that the small bowel biopsies proved most useful in the differential diagnosis of the various causes of steatorrhea. They reported a recognizable lesion in idiopathic steatorrhea. They were also able to follow the course or response to treatment of intestinal abnormalities by repeated suction biopsy.

Also in late 1959, Posey and Stephenson (35) published the very interesting results of intestinal biopsies using the Shiner biopsy tube on 4 subjects with steatorrhea. One of these was a woman with a history typical of non-tropical

sprue of 25 years duration; one was a case of childhood idiopathic steatorrhea persisting into adulthood; one of an obscure malabsorption state characterized by anemia, weight loss, abnormal Schilling test and abnormal Thorn test, and without obvious steatorrhea; and one case of idiopathic steatorrhea appearing in an older man. Although the therapy was not considered in this report, it was noted that only one patient, the individual with adult celiac disease, responded in any manner to a gluten-free diet. The other three required steroids in maintenance doses to be brought under control. It is difficult to reason that the four patients could have four different forms of the same disease despite the similarity of the biopsy findings. The researchers felt it more logical to assume that four unique etiologically unrelated disorders presented themselves in the final pathway of a malabsorption syndrome, one facet of which is mucosal atrophy.

The importance of the biopsy procedure, however, was not minimized in the report. The authors believed that the biopsy technique represented an important tool which could be of great use in obscure cases, and which is a short-cut in determining the presence of malabsorption whenever more elaborate procedures are unavailable or where the results of such tests are equivocal. They did not feel, however, that the method provides an etiologic or definitive diagnosis,

but rather should be looked upon as a means of establishing the presence of a general disorder, the malabsorption syndrome, which includes, as mentioned earlier in this paper, a variety of disparate diseases.

The findings which these four biopsy specimens presented were, briefly: mucosal atrophy with shortening, lateral fusion, and blunting of the villi, with an increase in mucus-choked goblet cells, and with a variable amount of submucosal edema and round-cell infiltration.

One of the most recent and complete reports correlating biopsy specimens with the diagnosis of idiopathic steatorrhea was published in 1960 by Cooke, et al (11). In an attempt to clarify the problem, the findings in the jejunal biopsies of 58 patients with "idiopathic steatorrhea" were presented and correlated with the clinical features, hematologic, radiographic and biochemical findings and the absorption of $Co^{58}$ vitamine $B_{12}$.

The 58 patients fulfilled the diagnostic criteria of "idiopathic steatorrhea" or "adult celiac disease." The patients were selected at random from those attending the steatorrhea clinic at the Birmingham (England) General Hospital. Biopsies were also performed on a control group of 65 persons of comparable age groups; normal medical students, patients with pernicious anemia, iron-deficiency anemia, steatorrhea due to pancreatic disease, regional

enteritis and enterocolitis, and miscellaneous disorders not associated with gastrointestinal disorders. Twenty biopsies were done with the Shiner biopsy tube, and the remaining 103 biopsies were obtained with the Crosby biopsy capsule.

The clinical and laboratory findings are summarized in the following table:

SUMMARY OF CLINICAL AND LABORATORY FINDINGS

| | Group I* | Group II** | | |
|---|---|---|---|---|
| | | A | B | C |
| Total | 27 | 12 | 8 | 7 |
| Age (yr.) at onset | 1-53 | 4-46 | 26-59 | 14-73 |
| Major symptoms: | | | | |
| A. diarrhea, malaise, weight loss | 26 | 12 | - | - |
| B. anemia | 1 | - | 8 | 7 |

(cont. over)

* Group I - Patients with "flat" jejunal biopsy
** Group II - Patients with "abnormal" villi in jejunum: A , patients in whom diarrhea, malaise, and weight loss were the major features; B, patients in whom severe anemia responding to Vitamin $B_{12}$ was the major feature; C, patients in whom severe anemia responding to folic acid was the major feature.

Figures in parenthesis refer to the total number in whom the test was performed.

SUMMARY (Continued)

| | Group I | Group II | | |
|---|---|---|---|---|
| | | A | B | C |
| Bone marrow: | | | | |
| A. megaloblastic | 4 | 2 | 5 | 7 |
| B. normoblastic | 3 | 1 | - | - |
| Initial Hbg. (gm.%) | 7.0-16.3 | 10.0-14.0 | 4.8-7.8 | 3.2-10.3 |
| Serum vitamin $B_{12}$, less than 105 g | 4(27) | 6(12) | 4(4) | 1(7) |
| Vitamin $B_{12}$ absorption | | | | |
| normal | 16(18) | 5(7) | 1(6) | 6(6) |
| Achlorhydria | 4(13) | 4(7) | 6(6) | 0(7) |
| Steatorrhea | 27 | 12 | 8 | 5(7) |
| Xylose absorption | | | | |
| abnormal | 10(10) | 4(7) | 3(4) | 6(7) |
| Folic acid excretion | | | | |
| test abnormal | 15(15) | 7(7) | 4(5) | 4(5) |
| Remission induced by | | | | |
| gluten-free diet | 8(11) | 6(9) | - | - |

The histologic findings in these 58 patients with "idiopathic steatorrhea" were classified into three groups.

In the 27 patients in Group I, the biopsies were strikingly uniform. The mucosa was of normal or reduced width, and the surface completely or almost completely flattened,

and no normal villi were present. When present, the villi appeared broad and short and tended to fuse together; elsewhere there was no suggestion of villus formation at all. The surface epithelium was grossly abnormal. The cell cytoplasm was often vaculated, the nuclei varied in size, shape and degree of staining. There were very few goblet cells in the surface epithelium, but they were present in the tubules. The glandular tubules were normal or reduced in number, and often elongated and dilated. In the lamina propria there was mild or moderately heavy cellular infiltration, consisting predominantly of plasma cells.

Although in 11 patients biopsy was performed during clinical remission and in 16 during the initial illness or during relapse, the histologic appearance was essentially the same.

In the 27 cases in Group II the microscopic appearances were considered to be abnormal, but to differ from the appearances seen in Group I. In particular, the absence of villi and flattening of the free surface were not found. In a few instances the mucosa was thicker than normal, although in most it was of approximately normal or reduced width. Villi were present in all specimens, but abnormal in size, shape, and structure; they were commonly short; many were broad and squat. The surface epithelium varied considerably in the degree of epithelial change, but this was never

so severe as seen in Group I. The glandular tubules showed little elongation or dilatation. Goblet cells were often numerous.

In the lamina propria there was mild, moderate, or heavy cellular infiltrate, but it was mostly of lymphocytes with less numerous plasma cells. The cellular infiltrate tended to be more unevenly distributed than in Group I, and was heavier in the deeper parts of the mucosa than in the villi themselves.

The 12 patients in whom diarrhea and malaise were main symptoms resembled the patients with the "flat" biopsies of Group I, but on more detailed analysis points of difference could be demonstrated in the majority.

There were 4 patients in Group III who presented with anemia and diarrhea to whom the diagnosis of idiopathic steatorrhea had been applied and in whom the jejunal biopsies proved to be normal. Further investigation brought out long histories of chronic iron loss; and upon iron therapy parenterally, the abdominal symptoms ceased.

As a result of these studies, the researchers concluded that there were at least two and probably three etiologically different disorders responsible for "idiopathic" nontropical steatorrhea. The most striking and homogenous histologic group in the series was Group I, with the "flat" jejunal mucosa, and this appears to represent an etiologic

group separate from Group II, with the "abnormal villi." Analysis of the clinical and biochemical features in the two groups as presented in Cooke's report, seem to support this belief.

As previously presented, Himes and Adlersberg considered the "flat" biopsies to be the end result of a pathologic lesion which in the earlier or less severe case would have an appearance similar to the "abnormal villi" group. Conversely, if regeneration would occur in the "flat" mucosal specimens, bizarre and abnormal villi would result. Cooke and his associates, however, in strengthening their beliefs, refer to the fact that 3 patients in Group II had the disease for 15 to 18 years, and that neither time nor lack of treatment appeared to have produced a "flat" biopsy.

On the other hand, 2 patients in Group I were biopsied at a time of relatively mild symptoms which had been present for only one year. Six patients in Group I had been in good health with no symptoms for periods of 5 to 10 years when the biopsies were performed, yet all 6 had "flat" biopsies, with absence of villi and a disordered epithelium. These and similar observations presented by Cooke's group suggest that regeneration does not occur to any significant degree, although complete clinical remission may occur.

The occurrence of virtually normal absorption of $Co^{58}$ vitamin $B_{12}$, the infrequent finding of low serum levels of

vitamin $B_{12}$, the relatively low incidence of severe anemia and of severe abdominal pain are further evidence that the disorder in the patients of Group I is a distinct entity.

In reference to patients of both groups responding to a gluten-free diet, Cooke refers to published case reports of patients with secondary steatorrhea who have been benefited by a gluten-free diet. From these reports, Cooke infers that the action of such a diet may be of a non-specific effect in some intestinal disorders and probably so acts on some patients with abnormal villi. However he does not infer that a gluten-free diet does not have a specific effect in celiac disease.

The histologic appearances in Group II are less uniform than those in Group I. Many have the characteristics of a chronic inflammation. In others the major change is in the appearance of the villi, with less severe epithelial disorder or cellular infiltration. It may well be that with many in the latter group, the small bowel lesion is a form of chronic jejunitis with various degrees of mucosal atrophy. It would have been interesting if Cooke had taken duodenal specimens on these patients since many reports show similar villi changes there also.

Jejunal biopsy appearances essentially similar to those described in Groups I and II have been described in this paper. At Children's Hospital, Birmingham (England), biopsies

have been performed on 26 children with steatorrhea, their ages from 2 to 14 years. Twenty-four who had the clinical features of idiopathic steatorrhea had histologic findings similar to Cooke's Group I. Cooke suggested that in the future the name idiopathic steatorrhea or adult celiac disease should be applied only to those patients with a "flat" jejunal biopsy.

The pathogenesis of the lesion of the "flat" jejunal biopsy is not known. The most prominent features are the absence of villi, the disordered epithelium with evidence of cell degeneration, and the predominantly plasma cell infiltration of the lamina propria. Evidence is accumulating that the basic defect in celiac disease is enzymatic or cellular and that the findings in the childhood idiopathic steatorrhea and the adult non-tropical sprue are identical; and that the latter is actually the adult manifestation of the childhood disease.

## SUMMARY

Beginning with Doctor Samuel Gee's classic and almost perfect description of celiac disease in 1888, the different disease entities which are included in the celiac syndrome were noted, including: (1) true or idiopathic celiac disease, also called the malabsorption syndrome; (2) celiac syndrome resulting from mechanical obstruction of digestion and/or absorption; (3) pancreatic insufficiency; (4) severe dietary deficiency; (5) gastrointestinal allergy; and (6) chronic intestinal infection.

Next, the non-specific laboratory findings helpful in the diagnosis of the celiac syndrome were mentioned. These included: (1) the high fat content of the stool, exceeding 5 gms. a day; (2) the lowered or flat absorption of vitamin A; (3) the lowered glucose tolerance test; (4) the lowered blood neutral fat and blood cholesterol levels; (5) the radiologic evidence of gross clumping of the barium; (6) the examination of the small bowel contents; (7) and the presence of a normochromic macrocytic or hypochromic anemia.

It was next noted that there is a group of infants who present the picture of celiac syndrome in which no clear-cut cause can be determined and the diagnosis is made largely by the process of exclusion. This group is considered to suffer from idiopathic celiac disease. A brief description of the specific findings in this disease entity is next made, including possibility of a family predisposition

and correlation to the incidence of diabetes mellitus. The important conclusions which relate the adult non-tropical sprue as representing a continuing manifestation of the childhood idiopathic celiac disease are included; and later in the paper this is again emphasized by the similarity of the biopsy findings in both diseases.

A thorough review of the literature concerning the pathologic findings in idiopathic steatorrhea from autopsy operative and biopsy specimens is next presented. This is traced back to Gee's own observations in which he was unable to demonstrate any conclusive findings. Other early observers noted atrophic changes in the small bowel; and as early as 1913, Justi described villi abnormalities from autopsy jejunal specimens. In 1922, Manson-Bahr studied eight cases of adult sprue and described villi changes and round-cell infiltration with absence of inflammatory processes.

Thaysen's monograph in 1932 concluded that all the previously reported changes were due to post-mortem autolysis and that there were no intestinal pathologic abnormalities in idiopathic steatorrhea. It was not until Paulley in 1954 disproved Thaysen's beliefs that interest in the pathology of the intestine was again stimulated.

The perfection of the intestinal biopsy tubes after numerous modifications of Wood's gastric tube, opened a new

field of investigation. With the ability of obtaining biopsy specimens from the small intestine with a minimum of discomfort to the patient, with minimal to no complications, and with fixation of the specimen in a few minutes, specimens could be studied under the light microscope with the complete elimination of autolytic changes.

In 1956, Shiner perfected the first of three biopsy tubes and felt that with such an aid, specimens obtained from the small bowel might show diagnostic changes in cases of steatorrhea. In 1957, Crosby perfected his intestinal biopsy capsule which he concluded to be safe, reliable, and technically uncomplicated. In 1959, the latest modification of the biopsy instruments was made by Rubin with his multipurpose suction biopsy tube which, because of its smaller diameter and greater flexibility, enabled him to use it with equal facility in infants, children, and adults. This tube was also able to biopsy multiple specimens with each intubation performed.

The remaining report reviews the findings of four series of biopsy and operative intestinal specimens of 15, 4, 8, and 58 patients diagnosed as having celiac disease and non-tropical sprue, all showing characteristic pathologic findings. Himes and Adlersberg, in their series of 15 patients, reported that the findings showed two stages of the disease. The early and mild stage showed changes in the

tips of the villi with thickening and clubbing, while further progression of the disease resulted in fusion of the villi and obliteration of the mucosal pattern resulting in an almost completely flat surface.

In Posey and Stephenson's study of four cases of steatorrhea (completely different forms of the disease), they concluded that their four unique and etiologically unrelated disorders had presented themselves in the final pathway of a malabsorption syndrome, one facet of which is mucosal atrophy. They felt the biopsy technique was an important tool in the diagnosis of steatorrhea, however it did not provide an etiologic or definitive diagnosis but rather showed the presence of a general disorder - the malabsorption syndrome.

The most recent and complete report on 58 patients with idiopathic steatorrhea biopsied by both the Shiner and the Crosby biopsy tubes was published by Cooke et al. in 1960. This series was carefully correlated with clinical, laboratory, and biochemical findings. Two distinct groups were found, one with "flat" biopsies and one with abnormal villi. These specimens, similar to Himes' and Adlersberg's description, were, on the basis of other findings, grouped into the true celiac disease with the "flat" biopsy and another unknown etiologic group with abnormal villi. On the basis of this work, Cooke felt that biopsy specimens could

be diagnostic of the celiac syndrome.

Throughout this survey, prominent researchers have reported that the intestinal biopsy tube represents a safe and reliable technique to obtain intestinal specimens. The complications and risks are at a minimum as compared to the diagnostic value of these specimens in many diseases of the gastrointestinal tract. Specimens can be successfully obtained in almost 90% of all attempts; and with the latest multipurpose tube, infants and children can be biopsied without undue risk. Also with the latter tube, multiple specimens can be obtained with one intubation. The benefit of these biopsied specimens for diagnosis and for evaluation of treatment has not yet been fully appreciated.

As for the use of such instruments as means for definitive diagnosis in celiac disease and non-tropical sprue, it is the opinion of this observer that when correlated with clinical and laboratory findings, such a procedure may well be diagnostic. This is especially true in those cases of childhood steatorrhea which have apparently been reported to show specific pathologic findings in the jejunum.

## CONCLUSIONS

The celiac syndrome is reviewed with special attention to its disease entities and to helpful diagnostic procedures. The syndrome of idiopathic celiac disease is next discussed, including its relation to the adult disease of non-tropical sprue.

A review of the literature concerning the pathologic findings in idiopathic celiac disease and non-tropical sprue is presented, along with conclusions as to the relationship of these findings with the etiology and diagnosis of this disease. Special attention is placed on the role of the peroral intestinal biopsy instruments, including the historical development of these instruments, their application to diagnosis of gastrointestinal disease, and their particular use as a diagnostic tool in idiopathic steatorrhea.

BIBLIOGRAPHY


1. Adlersberg, P., and Schein, J. Clinical and pathological studies in sprue, J.A.M.A. 134:1459, 1947.

2. Anderson, D. Cystic fibrosis of pancreas (vs. celiac disease), Amer. J. Dis. Child. 56:344, 1938.

3. Astalde, G., and Strosselli, E. Biopsy of normal intestine, Amer. J. Digest. Dis. 5:175, 1960.

4. Beneke, H. Verh. D. disch. path. Ges., 1910, p. 132. cited by: Shein, J. Syndrome of non-tropical sprue with hitherto undescribed lesions of the intestine, Gastroenterology 8:439, 1947.

5. Bolt, R. J., Pollard, M., and Standaert, L. Transoral small-bowel biopsy as an aid in the diagnosis of malabsorption states, New Engl. J. Med. 259:7, 1958.

6. Bossak, E. T., Wang, C. I., and Adlersberg, D. Clinical aspects of the malabsorption syndrome (idiopathic sprue): clinical observations in 94 patients, J. Mount Sinai Hosp. 24:296, 1957.

Brandborg, L. L., Rubin, C. E., and Quinton, W. E. A multipurpose instrument for suction biopsy of the esophagus, stomach, small bowel, and colon, Gastroenterology 37:1, 1959.

8. Butterfield, C. E., and Perez-Santiago, E. Pathological changes in jejunal biopsies from sprue patients, Amer. J. Digest. Dis. 2:659, 1958.

_____, Jejunal biopsies in sprue, Ann. Int. Med. 48:109, 1958.

10. Colcher, H., and Aldersberg, D. Management of patients with malabsorption syndrome, J. Mount Sinai Hosp. 24:380, 1957.

11. Cooke, W. T., Peeney, A. L. P., and Hawkins, C. F. Symptoms, signs and diagnostic features of idiopathic steatorrhea, Quart. J. Med. 22:59, 1953.

12. _____, What is idiopathic steatorrhea? Amer. J. Digest. Dis. 2:601, 1957.

13. Crosby, H. C., and Kugler, H. W. Intraluminal biopsy of the small bowel: the intestinal biopsy capsule, Amer. J. Digest. Dis. 2:236, 1957.

14. Culver, P. J., Benson, J. A., Strauss, E., and Jones, C. M. Observations on the malabsorption syndrome based on the use of absorption tests and biopsy of the small bowel, Gastroenterology 36:459, 1959.

15. Ebbs, J. H., Thompson, M., and Steen, W. D. Etiological factors in celiac disease, Amer. J. Dis. Child. 79:936, 1950.

16. Faber, K. Arch. f. verdauangs - Krankheiten 9:336, 1904; cited by Butterfield, C. E., and Perez-Santiago, E. op. cit. 8.

17. Fone, D. J., Cooke, W. T., Meynell, M. J., Brewer, D.B., Harris, E. L., and Cox, E. V. Jejunal biopsy in adult coeliac disease and allied disorders, Lancet 1:733, 1960.

18. Frazer, A. Present knowledge of celiac syndrome, J. Pediat. 57:262, 1957.

Gardner, F. H. Sprue. In: Principles of Internal Medicine, 3rd Ed., edited by T. R. Harrison, New York, McGraw-Hill Book Co., 1958. pp. 528-33.

20. Gee, Samuel. On the celiac affection, St. Bart. Reports, Vol. XXIV, 1888.

21. Gerrard, J. W., Astley, R., French, J. M. Coeliac Disease, Brit. J. Radiol. 25:526, 1952.

22. _____, Ross, C. A. C., Astley, R., French, J.M., and Smellie, J. M. Coeliac disease: Is there a natural recovery? Quart. J. Med. 24:23, 1955.

Glass, G. P. I. Intestinal absorption and hepatic uptake of vitamin $B_{12}$ in diseases of the gastro-intestinal tract, Gastroenterology 30:37, 1956.

Glazer, I., and Adlersberg, D. Volvulus of the colon; a complication of sprue, Gastroenterology 24:159, 1953.

Hawkins, C. F., Peeney, A. L. P., and Cooke, W. T. Refractory hypochromic anemia and steatorrhea: treatment with intravenous iron, Lancet, 2:387, 1950.

26. Himes, H. W., and Adlersberg, D. Pathological changes in the small bowel in idiopathic sprue; biopsy and autopsy findings, Gastroenterology 35:142, 1958.

_____, Pathological studies in idiopathic sprue, J. Mount Sinai Hosp. 24:251, 1957.

28. Justi, K. Arch. f. Schiffs u. Tropeor Hygiene 17:5, 1913; cited by: Butterfield, C. E., and Perez-Santiago, E. op. cit. 8.

29. Manson-Bahr, P. H. Sprue, Brit. Med. J. 2:17, 1914.

30. _____, The morbid anatomy and pathology of sprue, Lancet 1:1148, 1924.

Marshak, R. H., Wolf, B. S., and Elliasoph, J. Roentgen findings in the malabsorption syndrome, J. Mount Sinai Hosp. 24:362, 1957.

32. Parsons, L. G. Coeliac disease, Lancet 1:631, 1931.

Patterson, D., and McCreary, J. F. Celiac Syndrome, In: Pediatrics, Philadelphia, J. B. Lippincott Co., 1956. pp. 100-106.

Paulley, J. W. Observations on the etiology of idiopathic steatorrhea, Brit. Med. J. 2:1318, 1954.

Posey, E. L., and Stephenson, S. L. Peroral biopsy of duodenum and jejunum in the diagnosis of malabsorption syndrome, Southern Med. J. 52:691, 1959.

Rapoport, M. Celiac Disturbances. In: Textbook of Pediatrics, edited by W. E. Nelson and others, 7th Ed., Philadelphia, W. B. Saunders, 1959. pp. 721-26.

37. Sakula, J., and Shiner, M. Coeliac disease with atrophy of the small intestine mucosa, Lancet 2: 876, 1957.

Schein, J. Syndrome of non-tropical sprue, Gastroenterology 8:438, 1947.

39. Sheldon, W. Coeliac disease, Lancet 2:1097, 1955.

40. _____, Celiac Disease (Kenneth D. Blackfan Memo rialLecture), Pediatrics 23:132, 1959.

Shiner, M. Duodenal biopsy, Lancet 1:17, 1956.

_____, Jejunal biopsy tube, Lancet 1:85, 1956.

_____, Duodenal and jejunal biopsies: Discussion of method, its difficulties and applications, Gastroenterology 33:64, 1957.

_____, Small intestinal biopsies by the oral route, J. Mount Sinai Hosp. 24:273, 1957.

Smith, R. B. W., Spring, H., Crosby, W. H., and Sullivan, B. H. Peroral small bowel mucosal biopsy, Amer. J. Med. 25:391, 1958.

Stevenson, T. D., Little, J. A., and Langley, L. Pernicious anemia in childhood, New Engl. J. Med. 255:1219, 1956.

47. Still, G. F. On celiac disease, Lancet 2:163, 1918.

48. Sullivan, B. H., Spring, H., and Batzake, J. G. Peroral small bowel mucosal biopsy, J.A.M.A. 18: 2200, 1960.

49. Thaysen, T. E. H. Non-tropical Sprue, London, Oxford University Press, 1932. p. 232.

50. Thin, G. On a peculiar disease of hot climates, Practitioner 31:169, 1883.

51. _____. Brit. Med. J., June 14, 1890. As cited by Schein, J. op. cit. #38.

52. Tomenius, J. An instrument for gastric biopsies, Gastroenterology 15:498, 1950.

53. Volwiler, W. Gastrointestinal malabsorptive syndromes, Amer. J. Med. 23:350, 1957.

54. Weathered, B. Trans. Path. Soc. London 116:42, 1890. As cited by Schein, J. op. cit. #38.

Wood, I. J., Doig, R. K., Motteram, R., and Hughes, A. Gastric biopsy, Lancet 1:18, 1949.

Wood, I. J., Doig, R. K., Motteram, R., Weiden, S., and Moore, A. The relationship between the secretions of the gastric mucosa and its morphology as shown by biopsy specimens, Gastroenterology 12:949, 1949.