during these trials; that this dosage was effective in clearing most trophozoites of *P. falciparum* from the blood of the observed group by D+4 day, but that occasional parasites persisted; that *P. malariae* parasites, although persisting in the blood for more than seven days after treatment, do eventually disappear; that the effect of a single dose of B.W. 50-63, as assessed by parasite density indices, spleen rates, and average enlarged spleens, appears to persist for one month but to be lost during the next month.

During the preliminary trials no unexplained absence occurred among the 22 children treated with 25 mg. B.W. 50-63, and no complaint of headache, nausea, or vomiting was made by any of these children.

My thanks are due to Dr. S. L. A. Manuwa, O.B.E., Director of Medical Services, Nigeria, and to Dr. L. J. Bruce-Chwatt, Senior Malariologist i/c Malaria Service, Medical Department, Nigeria, for permission to publish this paper. I am also grateful to Brigadier J. S. K. Boyd, F.R.S., and Dr. L. G. Goodwin for providing supplies of B.W. 50-63, and for advance information on laboratory investigations.

REFERENCES

- Bruce-Chwatt, L. J. (1951). *Trans. roy. Soc. trop. Med. Hyg.*, **44**, 563.
 Falco, E. A., Hitchings, G. H., Russell, P. B., and Vander-Werff, H. (1949). *Nature, Lond.*, **164**, 107.
 — Goodwin, L. G., Hitchings, G. H., Rollo, I. M., and Russell, P. B. (1951). *Brit. J. Pharmacol.*, **6**, 187.
 Hackett, L. W. (1944). *J. nat. Malar. Soc.*, **3**, 121.
 W.H.O. Tech. Report Series No. 38 (1951). Malaria Conference in Equatorial Africa. Geneva.

VALUE OF GASTRIC BIOPSY IN THE STUDY OF CHRONIC GASTRITIS AND PERNICIOUS ANAEMIA*

BY

IAN J. WOOD, M.D., F.R.C.P.

Assistant Director, Walter and Eliza Hall Institute of Medical Research, Melbourne

I deeply appreciate the honour of being invited to deliver the Litchfield Lecture of 1951. It is the ambition of every Australian to visit Oxford, and I am most grateful for this additional privilege. I thought it would be of the greatest interest to you if I discussed the investigations we have been carrying out in Australia during the past three years on the gastric mucosa and its relationship to disease.

In 1946 Sir Macfarlane Burnet established a clinical research unit at the Walter and Eliza Hall Institute of Medical Research and the Royal Melbourne Hospital to work in the wards of the hospital and in the adjoining laboratories of the institute, and I have the privilege of being a member of this unit. We chose the alimentary canal for our studies, employing clinical observation, serial biochemical tests, x-ray examination, gastroscopy, and liver biopsy. We soon began to encounter a group of patients, mostly women between the ages of 35 and 60, who complained of long-standing relapsing flatulent dyspepsia, and examination of whom revealed diffuse epigastric tenderness, negative findings to a barium meal, persistent histamine-fast achlorhydria or hypochlorhydria, and rather inconclusive findings on gastroscopy. We were investigating one of these patients by test meal one day when a fragment of

tissue was sucked out through the Ryle's tube. On section, this proved to be gastric epithelium. With considerable trepidation we then set out to obtain fragments of gastric mucosa in order to correlate their histological structure with the symptoms, signs, and biochemical tests, just as we had been doing with some success with liver fragments obtained by needle biopsy.

The Biopsy Procedure

In collaboration with A. Hughes, of our institute, a flexible gastric biopsy tube has been developed during the past three years (Wood, Doig, Motteram, and Hughes, 1949; Wood, Doig, Motteram, Weiden, and Moore, 1949). It consists of a length of Bowden wire covered with plastic tubing to render it airtight. At the distal end is a metal cylinder housing a cylindrical knife, which can be moved up and down past a lateral hole by means of a wire which passes through the whole length of the instrument. The tube can be passed readily into the empty stomach. Negative pressure is exerted at the upper end, and this sucks in a knuckle of gastric mucosa through the lateral hole at the lower end. This knuckle of gastric mucosa is then cut off by pulling up the knife. The procedure is easily performed and causes little inconvenience to the patient. A fragment of mucosa 2 mm. in diameter and extending down to the muscularis mucosae is obtained. In the last 100 attempts at biopsy my colleague, R. K. Doig, has obtained fragments of gastric mucosa in 92.

The fragment is most frequently removed from the greater curvature of the body of the stomach. This has been confirmed by x-ray examination with the tube *in situ* and by gastroscopy carried out immediately after the biopsy, when the biopsy site can usually be seen. In this lecture I refer to fragments removed from the body of the stomach.

The complications following biopsy have been few. We have performed 552 biopsies on 462 patients and there has been no evidence of perforation or established ulceration. Haemorrhage in the form of haematemesis or melaena has occurred in six cases, but on no occasion has it seemed to threaten life.

In our unit Gunter has studied the lesion produced in the gastric mucosa of the cat when the same biopsy instrument is used. He found that there was rapid healing of the traumatized area. Two days after taking the fragment regeneration of the surface epithelium was seen, and it completely covered the area by the third day. By the eighth day almost complete healing had taken place, but acid- and pepsin-secreting cells were not identified.

The history of gastric biopsy is of interest. In 1894, Einhorn, of New York, collected fragments of gastric mucosa by vigorous aspiration with a stomach tube, and in 1906 Chevalier Jackson, of Philadelphia, skilfully carried out biopsy through his rigid oesophagoscope. Little attention was paid to his findings. It fell to the lot of two Americans—Kenamore, in 1946, and Benedict, in 1948—to draw attention to the value of the procedure, using an operating gastroscope.

Types of Lesion Investigated

I propose to discuss several patients seen in our unit who are representative of the three groups of diffuse gastric lesions which we have sorted out with the help of our pathologist, R. Motteram. I shall not discuss gastric ulcer or carcinoma, for here the mucosal changes

*An abstract of the Litchfield Lecture given at Oxford on June 6.

are not evenly spread; there is a zonal gastritis extending out from the lesion, giving variable findings. In duodenal ulcer, gastric biopsy of the body of the stomach usually shows a normal mucosa. It has been normal in 21 out of 29 cases. Of course, with duodenal ulcer there may be gastritis in the antrum.

We have found three main types of lesion in the gastric mucosa which, from a histological point of view, we had classified as superficial gastritis, atrophic gastritis, and gastric atrophy. We consider that superficial gastritis may resolve or may progress to atrophic gastritis, whereas once the stage of atrophic gastritis is reached there appears to be little or no tendency to resolution. The third group, gastric atrophy, shows minimal evidence of inflammatory reaction, there is gross atrophy of the acid- and pepsin-secreting cells, and the lesion is usually associated with pernicious anaemia and/or subacute combined degeneration of the cord.

Superficial Gastritis

Case 1.—An intelligent housewife aged 37 had suffered from intermittent flatulent dyspepsia for the past six years. Her symptoms began after taking large doses of aspirin for an attack of influenza. She suffered from intermittent attacks of burning epigastric pain and flatulence, coming on soon after taking food and relieved by alkali. She was a sensible but nervy woman and had considerable domestic worry. Her appetite was poor and her diet had been somewhat deficient, as she had many fads. Her symptoms were aggravated by periods of worry and subsided when she was on holiday. Examination showed slight atrophy of the tongue, diffuse epigastric tenderness, more pronounced during the exacerbations of symptoms, negative barium-meal test, and negative gastroscopy. There was no occult blood in the stools. We studied her over a period of three years with serial biopsies and test meals; the test meals always showed hypochlorhydria or achlorhydria and the biopsies superficial gastritis. During one acute attack she was admitted to the ward and treated with penicillin, administered by mouth, apparently with some benefit.

Case 2.—A housewife aged 43 suffered from flatulent dyspepsia for three months and then from biliary colic and jaundice. Operation revealed stones in the common duct and gall-bladder. These were removed, together with the gall-bladder. She remained well during the next three years except for slight flatulent dyspepsia, and on one occasion a gastro-duodenal haemorrhage with a fall in haemoglobin and with occult blood in the stools. Repeated test meals before and after operation showed histamine-fast achlorhydria. Gastric biopsy revealed superficial gastritis with diffuse superficial haemorrhage.

This finding of haemorrhage in the biopsy fragment was probably due to trauma at the time of taking the biopsy, but we have found it more often in the mucosa of patients suffering from gastritis than in the normal mucosa. It would appear that gastritis may lead to recurrent gastric bleeding—usually of minor degree, but sometimes more formidable. We believe that chronic gastritis may sometimes contribute to the anaemia seen in "Witts's anaemia." On rare occasions the haemorrhage may reach the severity encountered in bleeding peptic ulcer. It may then present a problem to the surgeon as well as to the physician.

Superficial gastritis appears to be associated with chronic flatulent dyspepsia which waxes and wanes, often influenced by worry, malnutrition, or alcoholism. Slow but persistent haemorrhage may occur. Superficial gastritis may resolve, especially if the causal factor, such as acute alcoholism, is removed. It is associated with hypochlorhydria or achlorhydria. If it resolves there is

improvement in the patient's symptoms, the level of the free acid, and the biopsy picture. It may progress to atrophic gastritis, with persistent achlorhydria and perhaps more pronounced symptoms.

Atrophic Gastritis

Atrophic gastritis would appear to be an advanced stage of superficial gastritis; here there is atrophy of the acid- and pepsin-secreting cells in addition to the inflammatory reaction.

Case 3.—A woman aged 62 suffered from flatulent dyspepsia for six years. Her diet was deficient. She was not an alcoholic. She had pain in the epigastrium immediately after taking food, which was often relieved by alkaline powder. Her appetite was poor and there was no vomiting. Her tongue was atrophic, cheilosis occurred at the corners of the mouth, and there was epigastric tenderness. Repeated test meals over two years showed persistent achlorhydria. Barium meal tests were negative and gastric biopsy showed severe atrophic gastritis. With an explanation of her condition, encouragement to take a good diet, and rest in bed during the acute exacerbations, she has maintained moderately good health. Serial biopsies over a two-year period showed no tendency to regeneration of the specific acid- and pepsin-secreting cells, and the inflammatory changes varied only slightly.

Case 4.—This patient, aged 64, had been a healthy young woman when she went to India in the Australian Nursing Service during the 1914-18 war. On the Northern Frontier she developed malaria and dysentery, and the food was of very poor quality. She returned to Australia two years later, wasted and weak, a shadow of her former self. She has never completely recovered. She has remained thin, with flatulent dyspepsia, hypochromic anaemia relieved by iron, koilonychia, constant histamine-fast achlorhydria, and often occult blood in the stools. Gastric biopsy showed atrophic gastritis. When given iron, encouragement to take a full diet, and the reassurance that she did not have cancer, she remained in moderately good health during the next three years. In fact, she has given great help to our unit by volunteering to be the subject of some of our studies. She has provided most valuable evidence that the mucosal lesion in chronic atrophic gastritis undergoes little change with the passage of years.

It is our belief that atrophic gastritis may cause recurrent flatulent dyspepsia, atrophy of the tongue, constant hypochlorhydria or more often achlorhydria, and be negative to a barium meal test. There may be hypochromic anaemia, due in part to blood loss from the mucosa, malnutrition, and impaired intestinal absorption of iron. There appears to be little or no evidence that pernicious anaemia or subacute combined degeneration of the cord occurs in this group. Neither is there any tendency to complete recovery except, perhaps, in the case of the irradiated stomach (see below).

Gastric Atrophy

Our studies of the gastric mucosa naturally led us to consider the histological picture in cases of pernicious anaemia and subacute combined degeneration of the cord (Doig, Motteram, Robertson, and Wood, 1950). We have found that, in pernicious anaemia, (1) there is severe atrophy of the gastric mucosa with a tendency to intestinal metaplasia; (2) there is complete or almost complete absence of the inflammatory changes seen in superficial gastritis and atrophic gastritis; (3) the mucosa shows no tendency to regenerate when given standard liver therapy over a period of years—one case was examined by Doig and Motteram after thirteen years of adequate liver therapy; and (4) the mucosa does not regenerate when given liberal weekly injections of

vitamin B₁₂. We have termed this lesion of pernicious anaemia and subacute combined degeneration of the cord "gastric atrophy," in contradistinction to superficial gastritis and atrophic gastritis.

Case 5.—A chocolate manufacturer aged 47 was found to be suffering from typical pernicious anaemia with haemoglobin of 7.5 g. which responded to intramuscular "campolon." Stomach biopsy was performed before treatment and after three, six, and fifteen months' treatment. He was then given a four-months course of treatment with 80 µg. of vitamin B₁₂ weekly. The biopsy was repeated. The gastric mucosa was found by Funder to have remained unchanged. Serial test-meal examinations showed histamine-fast achlorhydria.

What is the correlation between the histological findings and the clinical picture? Of 41 patients showing "gastric atrophy" in the gastric mucosa, 39 had pernicious anaemia and/or subacute combined degeneration of the cord. Thus we have two patients who perhaps may be expected to develop pernicious anaemia and/or subacute combined degeneration of the cord during the next few years. On the other hand, of 66 cases with "atrophic gastritis" only five had pernicious anaemia and/or subacute combined degeneration of the cord.

We have found three cases of probable subacute combined degeneration of the cord with a normal peripheral blood picture and bone marrow and with changes in the gastric mucosa identical with those of pernicious anaemia.

Moreover, we have found gastric biopsy to be of value in diagnosing those patients with possible subacute combined degeneration of the cord or pernicious anaemia who unfortunately have had treatment with liver before an adequate blood examination has been performed and whose nervous lesions are not entirely typical.

When does this gross atrophy of the gastric mucosa occur (Doig, Motteram, Weiden, and Wood, 1950; Doig, and Wood, 1950)? We do not know. Perhaps we have a slight clue. We did test meals on 134 apparently healthy medical students and found only one with persistent histamine-fast achlorhydria. This young man, whom we have called subject X, had suffered no dyspepsia. His mother had hypochlorhydria to gruel when aged 44, and his maternal grandmother had died of pernicious anaemia at the age of 70. Gastric biopsy showed considerable atrophy of the chief and parietal cells but with little inflammatory reaction. However, the change was not as pronounced as in pernicious anaemia. We shall watch his future with great interest.

Atrophic gastritis can be produced artificially by applying deep x-ray therapy to the gastric mucosa. This has been shown by Palmer, in America, and by Holman, in Tasmania, to be of benefit in some cases of duodenal ulcer with high gastric acidity. Thus, a textile worker, aged 36, with duodenal ulcer which did not respond to medical treatment had a normal gastric biopsy prior to irradiation. He was then given a three-weeks course of 2,000 r by Kaye-Scott. In our unit, Doig, Funder, and Weiden studied his progress in considerable detail over a period of 15 months and found that the inflammatory reaction and atrophy became pronounced, with the development of achlorhydria and greatly diminished pepsin secretion one month after the course of treatment (Doig, Funder, and Weiden, 1951). The mucosal changes remained thus for one month and then gradually

returned to a normal appearance with rising acid and pepsin secretion. Normal findings were observed after the lapse of one year. This close correlation between the test-meal findings and the appearance of the gastric mucosa is in keeping with our general experience (Wood, Doig, Motteram, Weiden, and Moore, 1949).

Conclusion

I should like to emphasize that in our opinion diffuse lesions of the mucosa of the body of the stomach occur and may produce symptoms. Superficial gastritis with flatulent dyspepsia may heal or progress to atrophic gastritis. Atrophic gastritis causes recurrent flatulent dyspepsia and shows little or no tendency to heal. Severe gastric atrophy without inflammatory changes is associated with pernicious anaemia and/or subacute combined degeneration of the cord, and is probably not the end-stage of chronic gastritis. It may well be a slow atrophic process of different aetiology. Much has to be learned of the aetiology, course, and prevention of superficial gastritis, atrophic gastritis, and gastric atrophy. If we discovered how to prevent gastric atrophy perhaps we could prevent the onset of pernicious anaemia or the more tragic disease, subacute combined degeneration of the cord.

REFERENCES

- Doig, R. K., Funder, J. F., and Weiden, S. (1951). *Med. J. Aust.*, **1**, 828.
 — Motteram, R., Robertson, E. G., and Wood, I. J. (1950). *Lancet*, **2**, 836.
 — Weiden, S., and Wood, I. J. (1950). *Ibid.*, **1**, 948.
 — and Wood, I. J. (1950). *Med. J. Aust.*, **2**, 565.
 Wood, I. J., Doig, R. K., Motteram, R., and Hughes, A. (1949). *Lancet*, **1**, 18.
 — Weiden, S., and Moore, A. (1949). *Gastroenterology*, **12**, 949.

VITAMIN B₁₂ IN MEGALOBlastic ANAEMIA OF PREGNANCY AND TROPICAL NUTRITIONAL MACROCYTIC ANAEMIA

BY

S. CHAUDHURI, M.B., M.R.C.P., D.C.H.

Professor of Medicine, Lady Hardinge Medical College,
New Delhi

The value of vitamin B₁₂ in Addisonian pernicious anaemia, subacute combined degeneration, nutritional macrocytic anaemia, and tropical macrocytic anaemia has been thoroughly acknowledged (West, 1948; Berk *et al.*, 1948; Ungley, 1948a; Hall and Campbell, 1948a, 1948b; Spies *et al.*, 1948a, 1948b; 1949; Patel, 1948, 1950; Das Gupta, 1950). Reports of its use in megaloblastic ("pernicious") anaemia of pregnancy, however, are contradictory. Ungley (1948b), Bethell *et al.* (1948), Day *et al.* (1949), and Das Gupta (1950) used it without any response, while Patel and Kocher (1950) reported its successful use in five cases of anaemia of pregnancy and the puerperium. Patel (1950) also reported that a higher dosage was required in tropical macrocytic anaemia and in megaloblastic anaemia of pregnancy than in pernicious anaemia. A report of 21 cases of megaloblastic anaemia in women patients admitted under my care at Lady Hardinge Hospital, New Delhi, and treated with vitamin B₁₂, is here presented.