obtained. Such changes as were seen—focal necrosis or portal infiltration—were found to an equal extent in the control animals of the same age which had received arachis oil only. Among the 68 rats dying during the course of the experiments liver necrosis characteristic of acute poisoning with D.D.T. was seen in two. This suggested that there had been an acute lowering of the animal's tolerance to D.D.T. owing to intercurrent infection. No rat showed any histological evidence that could be called characteristic of chronic D.D.T. poisoning. The findings of Fitzhugh and Nelson (1947) and Laug *et al.* (1950) have not been confirmed. Sections of normal rat liver were compared after fixation in 10% formalin, 10% formol saline, and Helly's fluid for various periods and staining with haematoxylin and eosin. Basophilic cytoplasmic granules are often observed in the normal rat liver cells. Margination of basophilic cytoplasmic granules are common fixation artifacts. They are seen especially in formalin-fixed tissues and are more marked if the tissue is deliberately badly fixed. Formalin is not a suitable fixative for cytological study. The photomicrographs that illustrate the paper of Laug *et al.* (1950) suggest badly fixed tissues.

Summary

Rats given 0.36, 3.6, and 36 mg. D.D.T. per kg. daily for up to 60 weeks showed no lesions that were characteristic of chronic poisoning with D.D.T. The incidence of naturally occurring pathological lesions was equally distributed among the rats receiving D.D.T. and among the control rats receiving arachis oil alone. No significant structural disturbance has been found in the kidneys and heart.

We gratefully acknowledge much assistance from Brigadier A. Sachs, Director of Pathology for the Army, and Mr. Solomon, of the Agricultural Research Council's staff.

REFERENCES

- Cameron, G. R., and Burgess, F. (1945). *British Medical Journal*, **1**, 865.
 Dunbar, P. B. (1949). *Food, Drug, Cosmetic Law Quart.*, p. 233.
 Fitzhugh, O. G., and Nelson, A. A. (1947). *J. Pharmacol.*, **89**, 18.
 Judah, J. D. (1949). *Brit. J. Pharmacol.*, **4**, 120.
 Laug, E. P., Kunze, F. M., and Prickett, C. S. (1951). *Arch. industr. Hyg.*, **3**, 245.
 — Nelson, A. A., Fitzhugh, O. G., and Kunze, F. M. (1950). *J. Pharmacol.*, **98**, 268.

Replies to a questionnaire on student health sent out to universities in 53 countries had been received by the International Association of University Professors and Lecturers from 23 countries at the time of the Basle conference in 1949. The Association had decided at a previous conference that the universities should undertake a fuller responsibility for the health of their students, and recommended that arrangements should be made to examine students both before and regularly after entry. The replies made it quite clear that importance is generally attached to chest examinations; on the other hand, most universities are against any exacting psychological examination of the ordinary student's mental health, and an indirect approach is favoured. Prague uses intelligence tests as part of the entrance examination. Out of 91 universities having student health schemes, only about 40 provide physical education for those who do not take part in athletics. Closer supervision of lodgings and hostels is advocated, and the importance of adequate nourishment is stressed. Instruction in personal and social hygiene is also advised, and the establishment of suitable amenities for looking after students who are ill; some universities have sick wards in their residence, others utilize the State health service. Full details of the conditions existing in various universities are published in *Communication 16, Report on Student Health Services* (I.A.U.P.L., London, W.C.2, price 1s.).

PRELIMINARY FIELD TRIALS ON A NEW SCHIZONTICIDE

BY

H. M. ARCHIBALD, M.B.E., M.B., Ch.B.

Malaria Service, Medical Department, Nigeria

"B.W. 50-63" is one of a series of 2,4-diamino-pyrimidines, being 2,4-diamino 5-*p*-chlorophenyl 6-ethyl pyrimidine. Certain similarities which this compound and proguanil showed in their antagonistic action on pteroyl-glutamic acid suggested that it might have antimalarial activity (Falco *et al.*, 1949), and this was shown to be the case (Falco *et al.*, 1951).

At the time these field trials started, laboratory experiments had shown that this drug was a successful schizonticide against *Plasmodium gallinaceum* in chicks, *P. cynomolgi* in monkeys, and *P. berghei* in mice. Toxicity tests on chicks had shown that, weight for weight, the drug had a toxicity similar to that of proguanil, but as its effective dose promised to be small its active dose would be safer in practice. B.W. 50-63 was 60 times as active as proguanil against *P. gallinaceum* and 200 times as active against *P. berghei*. The drug was also active against the blood-forms of *P. cynomolgi* in monkeys, but had no pronounced action upon the exoerythrocytic stages (Falco *et al.*, 1951). It was decided to test this drug in accordance with the method introduced by Bruce-Chwatt (1951) and recommended at the World Health Organization's Conference on Malaria in Africa (1951). A number of African children naturally infected with malaria, of age group 5-10 years inclusive, were used, drawn from schools in the vicinity of Lagos.

Observations of splenic enlargement were made on standing children by a seated observer, and the results were classified in accordance with Hackett's classification (Hackett, 1944). Parasite rates and densities were calculated from observations made on thick films, "negatives" being determined only after the examination of 200 thick-film fields. Parasite density counts were made by determining the parasite/leucocyte ratio from the thick film and calculating from that the number of parasites per c.mm. The rates recorded are those generally accepted. The indices recorded are:

1. *The average enlarged spleen*: an index of the size of splenic enlargements in a group. It is obtained by multiplying the number of individuals in each spleen size group (Hackett's classification) by the group number, and dividing the sum of these products by the total number of those examined who are found to have enlarged spleens.
2. *The parasite density index*: an index of the density of parasitaemia in a group. It is obtained by multiplying the number of individuals in each parasite density group (Bruce-Chwatt, 1951) by the arbitrary group number, and dividing the sum of those products by the total number of individuals examined.

A preliminary test was carried out on a selected group of 12 indigenous schoolchildren, from a suburb of Lagos, whose bloods showed the presence of numerous *P. falciparum* trophozoites on D-1 and D day. On D day a single tablet of 25 mg. of the drug was taken by each child by mouth in the presence of the examiner. This preliminary test was carried out against control groups of untreated children and of children treated with 300 mg. proguanil in a single dose, orally,

on D day. The results of this preliminary test are given in Table I. They show: (i) that the drug was able to clear the blood of *P. falciparum* trophozoites by D+4 day; (ii) that neither *P. falciparum* gametocytes nor *P. malariae* were eliminated by B.W. 50-63 by D+7 day. A continuation of the trials appeared justified.

The next step was to discover if B.W.50-63 in this dosage cleared the blood any earlier than D+4 day, as a guide to the possible value of the drug as a "curative" as compared with its action as a "suppressive." Observations to this end were made on further groups of schoolchildren, selected as in the first test, and these observations were again made in comparison with controls as in the earlier investigation. An additional control group treated with 300 mg. mepacrine, given in a single oral dose on D day, was also included in this test, in view of the accepted place of mepacrine as a

quick-acting schizonticide. Data were collected on D, D+1, D+2, D+3, and D+4 days. The results are shown in Table II. They confirmed the findings of the first test, but suggest that B.W. 50-63 will clear the blood of *P. falciparum* trophozoites by D+3. The mepacrine control group, however, showed that mepacrine had an earlier effect.

In view of the results of these trials it was decided to investigate the persistence of B.W. 50-63 and its action when given in much lower doses than those tested before. Groups of schoolchildren, selected as already described, were tested against single oral doses on D day of (i) 10 mg. B.W. 50-63 and (ii) 5 mg. B.W. 50-63.

Observations were made on D, D+4, D+7, D+14, D+30, and D+ (60±15). The results are shown in Table III. They demonstrate that 5 mg. of the drug was quite as effective as many of the larger doses given

TABLE I.—Preliminary Trials of B.W. 50-63

	Untreated Controls Spleen Rate on D Day 7/12 = 58% Average Enlarged Spleen on D Day 15/7 = 2.1					Group Treated with 300 mg. Proguanil Spleen Rate on D Day 5/13 = 38% Average Enlarged Spleen on D Day 12/5 = 2.4					Group Treated with 25 mg. B.W. 50-63 Spleen Rate on D Day 11/12 = 92% Average Enlarged Spleen on D Day 25/11 = 2.3				
	-D	D	D+1	D+4	D+7	-D	D	D+1	D+4	D+7	-D	D	D+1	D+4	D+7
No. examined	12	11	12	12	10	13	13	12	13	13	12	12	12	12	12
No. absent	—	1	0	0	2	—	0	1	0	0	0	0	0	0	0
Crude parasite rate	12/12	10/11	9/12	11/12	9/10	13/13	12/13	10/12	2/13	3/13	12/12	12/12	12/12	6/12	5/12
<i>P. falciparum</i> trophozoite rate	100-0	91-0	75-0	91-6	90-0	100-0	92-0	83-5	15-4	23-0	100-0	100-0	100-0	50-0	41-6
<i>P. falciparum</i> gametocyte rate	12/12	10/11	9/12	10/12	9/10	13/13	12/13	9/12	0/13	0/13	12/12	12/12	12/12	0/12	0/12
<i>P. malariae</i> rate	100-0	91-0	75-0	91-6	90-0	100-0	92-0	75-0	0	0	100-0	100-0	100-0	0	0
P.D.I.	1/12	2/11	2/12	4/12	3/10	2/13	2/13	4/12	2/13	3/13	1/12	1/12	5/12	6/12	5/12
	8-3	18-1	16-7	33-3	30-0	15-4	15-4	33-0	15-4	23-0	8-3	8-3	41-6	50-0	41-6
	1/12	1/11	0/12	1/12	1/10	2/13	1/13	2/12	1/13	0/13	0/12	1/12	1/12	1/12	1/12
	8-3	9-1	0	8-3	10-0	15-4	7-7	16-7	7-7	0	0	8-3	8-3	8-3	8-3
	47/12	35/11	18/12	24/12	28/10	47/13	49/13	27/12	3/13	3/13	55/12	38/12	35/12	8/12	6/12
	3-9	3-2	1-5	2-0	2-8	3-6	3-8	2-3	0-2	0-2	4-6	3-2	2-9	0-75	0-5

TABLE II.—Tests of the Possible Value of B.W. 50-63 as a "Curative"

	Untreated Controls Spleen Rate on D Day 2/5 = 40% A.E.S. on D Day 4/2 = 2					Group Treated with 300 mg. Proguanil Spleen Rate on D Day 14/16 = 87.5% A.E.S. on D Day 30/14 = 2.1					Group Treated with 300 mg. Mepacrine Spleen Rate on D Day 13/14 = 93% A.E.S. on D Day 36/13 = 2.6					Group Treated with 25 mg. B.W. 50-63 Spleen Rate on D Day 9/10 = 90% A.E.S. on D Day 18/9 = 2				
	D	D+1	D+2	D+3	D+4	D	D+1	D+2	D+3	D+4	D	D+1	D+2	D+3	D+4	D	D+1	D+2	D+3	D+4
No. examined	5	4	4	4	4	16	14	16	14	16	14	13	12	13	12	10	10	9	10	10
No. absent	0	1	1	1	1	0	2	0	2	0	—	1	2	1	2	0	0	1	0	0
Crude parasite rate	5/5	4/4	4/4	3/4	4/4	16/16	13/14	10/16	7/14	4/16	14/14	9/13	1/12	5/13	1/12	10/10	9/10	4/9	5/10	4/10
<i>P. falciparum</i> trophozoite rate	100-0	100-0	100-0	75-0	100-0	100-0	93-0	62-5	50-0	25-0	100-0	69-0	8-3	38-5	8-3	100-0	90-0	44-4	50-0	40-0
<i>P. falciparum</i> gametocyte rate	5/5	4/4	4/4	2/4	3/4	15/16	12/14	1/16	1/14	0/16	12/14	5/13	0/12	1/13	0/12	9/10	8/10	1/9	0/10	0/10
<i>P. malariae</i> rate	100-0	100-0	100-0	50-0	75-0	94-0	86-0	6-25	7-1	0	85-75	38-5	0	7-7	0	90-0	80-0	11-1	0	0
P.D.I.	1/5	0/4	2/4	2/4	1/4	2/16	2/14	5/16	3/14	3/16	1/14	4/13	1/12	5/13	1/12	2/10	4/10	2/9	4/10	3/10
	20-0	0	50-0	50-0	25-0	12-5	14-3	31-3	21-4	18-75	7-1	30-8	8-3	38-5	8-3	20-0	40-0	22-2	40-0	30-0
	1/5	2/4	1/4	0/4	0/4	6/16	6/14	7/16	4/14	1/16	4/14	1/13	0/12	1/13	0/12	1/10	4/10	2/9	1/10	1/10
	20-0	50-0	25-0	0	0	37-5	43-0	43-75	28-6	6-25	28-6	7-7	0	7-7	0	10-0	40-0	22-2	10-0	10-0
	—	—	—	—	—	—	—	—	—	—	1/14	1/13	0/12	1/13	0/12	1/10	1/10	0/9	1/10	1/10
	16/5	11/4	13/4	5/4	11/4	55/16	43/14	21/16	8/14	4/16	60/14	17/13	1/12	7/13	1/12	32/10	25/10	6/9	6/10	4/10
	3-2	2-8	3-25	1-25	2-8	3-4	3-1	1-3	0-57	0-25	4-3	1-3	0-1	0-5	0-1	3-2	2-5	0-7	0-6	0-4

TABLE III.—Tests of the Persistence of the Action of B.W. 50-63

	Group Treated with 10 mg. B.W. 50-63						Group Treated with 5 mg. B.W. 50-63					
	D	D+4	D+7	D+14	D+30	D+60	D	D+4	D+7	D+14	D+30	D+60
No. examined	34	29	30	30	11*	24	48	28*	44	40	27*	31
No. absent	0	5	4	4	6	10	0	3	4	8	4	17
Spleen rate	24/32	—	—	13/28	6/11	4/14	34/45	—	—	14/40	10/27	13/30
A.E.S.	75-0	46-5	54-5	28-6	28-6	28-6	75-6	—	—	35-0	37-0	43-0
	48/24	—	—	25/13	10/6	7/4	69/34	—	—	22/14	14/10	23/13
Crude parasite rate	34/34	12/29	3/30	2/30	1/11	11/24	48/48	5/28	11/44	1/40	1/27	11/31
<i>P. falciparum</i> trophozoite rate	100-0	35-5	10-0	6-6	9-1	46-0	100-0	17-9	25-0	2-5	3-7	35-0
<i>P. falciparum</i> gametocyte rate	30/34	2/29	1/30	1/30	1/11	10/24	45/48	0/28	1/44	0/40	0/27	11/31
<i>P. malariae</i> rate	88-0	6-9	3-3	3-3	9-1	41-6	93-8	0	2-3	0	0	35-0
	10/34	7/29	1/30	2/30	0/11	3/24	6/48	4/28	11/44	1/40	1/27	0/31
	29-4	24-2	3-3	6-6	0	12-5	12-5	14-3	25-0	2-5	3-7	0
	8/34	6/29	2/30	0/30	0/11	0/24	8/48	3/28	1/44	0/40	0/27	1/31
	23-6	20-7	6-7	0	0	0	16-7	10-7	2-3	0	0	3-2
P.D.I.	125/34	17/29	3/30	5/30	1/11	42/24	183/48	7/28	11/44	1/40	1/27	33/31
	3-7	0-6	0-1	0-2	0-09	1-75	3-8	0-25	0-25	0-03	0-04	1-1

* Only one school available.

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.