VOL. IV NO. 1 JULY-SEPTEMBER

Acta Medica Philippina

Published Four Times a Year
by the
COLLEGE OF MEDICINE
and the
INSTITUTE OF HYGIENE

MANILA
PHILIPPINES
1947

ACTA MEDICA PHILIPPINA

Published by the College of Medicine and the Institute of Hygiene, University of the Philippines

Entered as second class matter at the Manila Post Office on October 5, 1939

ACTA MEDICA PHILIPPINA

Publication Office: College of Medicine, University of the Philippines
Manila

VOL. IV JULY-SEPTEMBER NO. 1

Note

As a result of World War II, publication of the ACTA MEDICA PHILIPPINA was suspended in 1941. The last number was Volume III, Number 1, July-September, 1941. Volume III, therefore, has only one number.

With this issue, Volume IV, Number 1, we resume publication of the ACTA.

THE EDITORS

IN MEMORIAM

CANDIDO M. AFRICA, M.D.

On February 12, 1945, Doctor Africa, a civilian and non-combatant, was shot in cold blood by Japanese soldiers, for reasons known only to themselves.

Born in Lipa, Batangas, on October 2, 1895, Doctor Africa was a product of the Philippine public schools. Soon after his graduation from the College of Medicine, U. P. in 1920, he was appointed instructor in Parasitology and Tropical Medicine. In 1939 he became full Professor and Head of the Department of Parasitology, a position he held until his death. From 1928-1931 he was a Fellow of the University of the Philippines and later Research Fellow of the Rockefeller Foundation. In 1929 he received his Diploma in Tropical Medicine and Hygiene from the London School of Tropical Medicine and Hygiene, and, before returning, visited other foreign institutions.

He became a Fellow of the Royal Society of Tropical Medicine and Hygiene in England and was member of the American Society of Parasitologists; the National German Association of Tropical Medicine; the Helminthological Society of Washington, D. C.; Phi Kappa Phi; and the Philippine Scientific Society; He was Secretary-Treasurer of the Philippine Medical Association in 1940-41, Charter Member of the National Research Council, and the first

Managing Editor of the Acta Medica Philippina. In 1935, he was selected Distinguished Alumnus of the University of the Philippines, thus automatically becoming a member of the Board of Citizens. He represented his country as delegate to the Third International Congress of Microbiology in 1939.

His contributions to Parasitology and Tropical Medicine number sixty-six and his papers * were published in all leading medical journals everywhere in the world, including Japan. Outstanding among these contributions was his discovery of the role of Heterophyids in heart failure, a monograph published by the Acta Medica Philippina in 1940. When the war broke out, he was completing another important work, the demonstration of exo-erythocytic-like bodies in certain cases of human malaria.

At the opening session of the Thirty-ninth Annual Convention of the Philippine Medical Association, a gold medal and diploma with citation were posthumously awarded to Doctor Africa. The last part of the citation reads:

"for his many other investigations in the field of parasitology which revealed the breadth and depth of his interest and enlarged the frontiers of knowledge in his field — all brought him international renown as a leader in his line and forward recognition of the Filipino scientist abroad, as has not been achieved before, thus reflecting glory and honor upon his calling and country."

---

* A complete list compiled by Dr. T. P. Pesigan was published in the Journal of the Philippine Medical Association 22: 218, (May) 1946.

INCIDENCE OF HEART DISEASE AMONG FILIPINOS A STATISTICAL STUDY BASED ON 10,437 AUTOPSIES

by

AGERICO B. M. SISON, M.D., VICENTE B. JIMENEZ, M.D.,
and VIRGINIA S. RODRIGUEZ, M.D.
Dept. of Medicine, College of Medicine,
University of the Philippines and the
Philippine General Hospital

MATERIAL—This series covers the period from 1932-1941 and is based on the clinical and autopsy records of patients from the different wards of the Philippine General Hospital. These records were totally destroyed by enemy action during the Battle of Manila.

DISTRIBUTION: In a total of 10,437 autopsies, from 1932-1941, 1,464 were cardiac cases. Of these, 55.6% were males and 44.4% were females. The yearly average of cardiac cases was 14%. The incidence of the various forms of heart disease is shown in Table I.

Since 1939, cardiac cases have been considered as a group; more detailed and extensive studies, especially with reference to etiology, age, sex, symptomatology, electrocardiography, etc., were carried out. Table II shows the age incidence.

CRITERIA FOR GROSS STRUCTURAL DIAGNOSIS:*

I. Arteriosclerotic Heart Disease:

1. Coronary Arteriosclerosis of all degrees varying from intimal sclerosis to atherosclerosis with calcification and/or erosion.
2. Presence of recent or old thrombi partially or completely occluding the lumina of the coronary vessels.
3. Presence of myocardial infarction to the exclusion of other extra-cardiac causes of infarcts, e.g., trauma.
4. Obvious changes affecting the myocardium in the form of fibrosis which may acquire a diffuse or a limited distribution.

* Modified from "Criteria for Diagnosis" of the New York Heart Association.

5. The size of the heart is not greatly altered and may even be relatively small unless passively dilated.
6. There may be a ventricular aneurysm, the result of a previous myocardial infarction.

II. Hypertensive Heart Disease:

1. Cardiac hypertrophy and dilatation particularly of the left ventricle associated with any of the following conditions known to give rise to hypertension:
   a. Chronic nephritis (glomerular or arteriolosclerotic)
   b. Polycystic kidneys.
   c. Coarctation of the aorta.
   d Chronic pyelonephritis.
   e. Prolonged obstruction in the urinary passages, etc.
2. Absence of true myocarditis or true myocardial degeneration in spite of the hypertrophied muscle fibers.
3. Dilated aorta.
4. Normal valves.
5. Presence of known hypertensive condition before death.

III. Arteriosclerotic and Hypertensive Heart Disease:

The diagnosis rests upon the association of the conditions enumerated in both I and II.

IV. Rheumatic Heart Disease:

1. Presence of verrucose vegetations with tendency to affect particularly the mitral cusps at the line of closure with frequent development of mitral stenosis.
2. Endocarditis with subsequent scarring involving particularly the wall of the left auricle (auriculitis) just above the valves.
3. Occasionally, the presence of fibrinous or sero-fibrinous pericarditis.
4. Rarely, thrombotic occlusion of the smaller coronaries or "Rheumatic Arteritis". Coronary arteriosclerosis must be excluded.
5. Dilated and enlarged left auricle and right ventricle.
6. Dilated pulmonary artery.
7. Chronic passive congestion, etc.

V. Bacterial Endocarditis:

1. Presence of vegetations in the valves or endocardium (consisting of fibrin, leucocytes, red cells, bacteria, etc.)

2. Evidences of an antecedent rheumatic valvulitis or endocarditis or a congenital lesion in the heart.
3. Petechial hemorrhages in the skin and mucus membranes.
4. Secondary anemia.
5. Enlarged spleen, cloudy swelling of the viscera, etc.

VI. Infantile and Adult Beriberi Heart:

1. Dilatation and hypertrophy of the right side of the heart.
2. Prominent trabeculae and papillary muscles of the right ventricle.
3. Normal valves.
4. Chronic passive congestion and anasarca.
5. Absence of other findings to account for the death.

VII. Cor Pulmonale:

1. Right-sided hypertrophy and dilatation (with or without left ventricular enlargement) association with the
2. Presence of a *primary* lung condition known to be able to give rise to increased pressure in the pulmonary circuit:
   a. Emphysema.
   b. Bronchiectasis.
   c. Obliterative pleuritis.
   d. Extensive fibrosis, adhesions, etc.
3. Spinal deformities.
4. Pulmonary embolism or thrombosis.
5. Dilated pulmonary artery.
6. Chronic passive congestion, etc.

VIII. Syphilitic Heart Disease:

1. Aortitis with dilatation, and atheromatous deposits in the intima.
2. Longitudinal wrinkling or ridging of the intima with often a sharp demarcation of the extent of the lesion in the thoracic aorta.
3. Mesaortitis.
4. Aortic insufficiency.
5. Cardiac hypertrophy especially of the left ventricle.
6. Presence of an aortic aneurysm.

7. Frequently, obstruction of the orifices of the coronary arteries (Coronary Ostial Stenosis).
8. Rarely, myocardial gummata.

IX. Congenital Heart Disease:

The diagnosis of congenital cardiac conditions rests upon the presence of abnormalities in the anatomical structure of the heart obviously dating from birth (Abbott).

COMMENTS

Many textbooks still teach that hypertension and rheumatic infection are seldom seen in the tropics. We hope this study will correct this impression. In the Philippines, at least, these two conditions are the most important etiologic agents in the causation of heart disease. Thus for a period of 10 years, 1932-1941, rheumatic heart disease comprised 3.4% of deaths from all causes and 24.7% of all cardiac cases. Hypertension was not far behind and actually, from 1939 to the first half of 1942, surpassed rheumatic infection, being found in 3.8% of all autopsies. More surprising than the total number of cases is the trend, which has been going up since 1932.

Comparison with the statistics of other workers in temperate countries points up the need of revision of the belief that these two diseases are seldom found in the tropics. Maher, Sittler and Elliott in Chicago reported an incidence of 50.3% for arteriosclerosis and hypertension; Coffen in Portland gave 56.3% in 1929; Stone, Vanzant, 61.4%; Schwab and Schultze, 77.4% in 1931; Hedley in Washington, D.C., 61.4% in 1935 and White and Jones in New England, 64.9% in 1928. For rheumatic heart disease, the difference beween our incidence and that of other workers is much smaller. Maher and Sittler reported 29.3% in Chicago, Cabot in Boston reported 39.5% and Hamilton and Hallisey, 46.4%.

Syphilis as a cause of heart disease in the Philippines is infrequent and has remained steadily so throughout the period of this study. Thus while Flaxman in Cook County Hospital report 10.9%; Stone Vanzant, 19.3% in Galveston; Schwab and Shultze, 12%; Hedley, 12% in Washington and Glazer, 14% in Cincinnati, our average for the ten-year period is 1.8%.

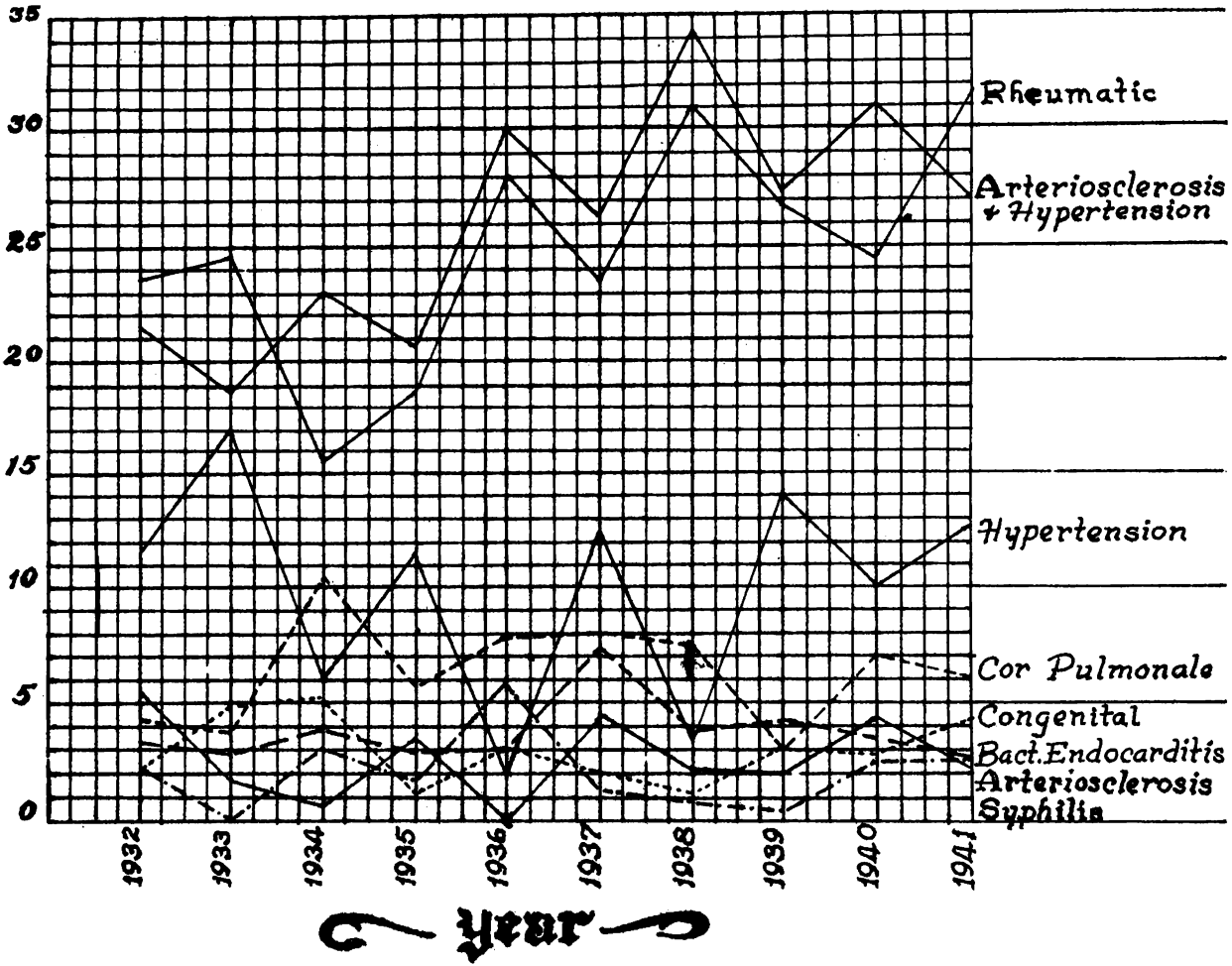

35
30
25
20
15
10
5
0
1932
1933
1934
1935
1936
1937
1938
1939
1940
1941
year
Rheumatic
Arteriosclerosis + Hypertension
Hypertension
Cor Pulmonale
Congenital
Bact. Endocarditis
Arteriosclerosis
Syphilis

TABLE I

INCIDENCE OF HEART DISEASE

Year	Arterio-sclero-sis and Hyper-tension		Arteri-oscleros-sis		Hyper-tension		Rheu-matic		Bacte-rial En docardi-tis		Infan tile Beribe-		Adult Beriber		Cor Pulmo nale		Syphilis		Congen ital		Various		Unde-ter-mined		Total	
	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F
1932	12	8	2	3	6	5	6	16	2	1	4	6	3	4	4	0	2	0	1	1	0	0	5	1	47	45
1933	9	11	1	1	10	8	10	16	1	2	9	10	0	2	3	1	0	0	2	3	0	1	3	3	48	58
1934	21	10	1	0	5	3	13	8	1	4	21	17	3	2	8	6	0	4	3	4	0	0	1	0	77	58
1935	23	15	5	1	12	8	19	14	2	3	29	14	4	4	8	4	1	2	2	1	0	0	7	0	112	66
1936	20	11	0	0	2	0	13	16	3	0	8	6	1	1	1	7	3	3	2	1	0	1	2	2	55	48
1937	24	15	5	2	9	10	16	19	6	5	10	6	1	0	6	6	1	1	2	1	0	0	2	2	82	67
1938	31	24	3	2	3	4	20	30	3	3	5	4	2	2	6	6	1	0	1	1	1	0	6	2	82	78
1939	36	18	5	1	21	6	19	33	5	3	11	8	1	1	5	3	1	0	5	3	1	1	5	2	115	79
1940	47	16	4	5	14	6	23	26	3	4	8	3	2	2	11	3	3	2	6	2	0	2	4	6	125	77
1941	30	9	1	2	8	10	13	32	3	1	4	4	1	1	5	4	3	0	4	2	1	4	0	2	73	71
1942*	10	4	4	1	0	2	7	8	0	2	0	0	0	0	0	2	0	1	0	2	0	1	0	0	21	23
Total	263	141	31	18	90	62	159	218	29	28	109	78	18	19	57	42	15	13	28	21	3	10	35	20	837	670

* First 6 months only.

TABLE II

AGE INCIDENCE OF HEART DISEASE FOR THE YEARS 1939, 1940, 1941 AND FIRST HALF OF 1942.

	Arterio-sclerosis and Hyper-tension	Arterio-sclerosis	Hyper-tension	Rheuma-tic	Bact. Endocar-ditis	Infan-tile Beriberi	Adult Beriberi	Cor. Pul-monale	Syphilis	Congen-ital
Below 1 yr.				1		29				6
1-10				15	2	9		1	1	8
11-20			4	46	3			5		5
21-30	17		7	38	5		4	6		1
31-40	10	1	11	38	6		1	6	3	2
41-50	36	6	12	16	4		2	4	4	
51-60	53	9	11	7			1	7	2	
61-70	40	5	16					2		
Over 70	23	2	4							2

INFANTILE BERIBERI IN THE PHILIPPINES

JOSE ALBERT, M.D.* and MOISES B. ABAD, M.D.
Department of Pediatrics, College of Medicine, University of the Philippines, and the Philippine General Hospital

There are problems of beriberi in general and of infantile beriberi in particular which are still unclear, and probably will remain so for a long time, since "medicine is not, and never will be, an exact science." In this report, we have restated some of the most important of these problems, in the hope that by considering them against the background of data gathered from 968 cases of infantile beriberi seen in the Pediatrics Ward of the Philippine General Hospital from 1914-1946, possible solutions or fresh approaches might become apparent.

Etiology.—Although beriberi was known to the Chinese as early as 2600 B. C. and to the Romans as early as 24 B. C.(2), its etiology is not settled. The prevailing opinion is that it is a nutritional deficiency disease. Until recently we accepted this opinion and believed that Vitamin B_1 was the deficient factor. However, recent studies have forced us to modify this opinion.

a. Race.—In 1931, Albert reported a series of 500 cases of infantile beriberi in which he pointed out that the condition was not seen among Caucasians or among the offspring of Caucasian-Malayan parents. The explanation given was the difference in diet and social scale; but low-income families, where the diet is obviously inadequate, are found in all groups—Caucasian, mixed, and Malayan.

b. Age.—Infantile beriberi, as the term implies, occurs almost exclusively during the first year of life. As can be seen in Table 3 the most dangerous period is 1-4 months. During this time, the disease is most frequent as well as more severe. Guerrero and Quintos(9) stated that the greater number belong to the group between one and three months old. Beyond the age of one year we do not see the disease. Why does beriberi affect only infants and adults and spare young children? Obviously, a thiamin-deficient diet cannot be the only answer.

---

* Doctor Albert died on August 13, 1946.

c. Monthly Variations.—Humidity, rainfall, temperature, atmospheric pressure probably exert some influence, for although infantile beriberi is found throughout the year, the incidence is significantly lower during the months of May and June and highest during November, December and January (Table 2).

d. Yearly Variations.—In May, 1928, Albert and Ocampo(3) published a paper wherein they voiced the opinion that "infantile beriberi was disappearing at least in Manila and its environs." They attributed this success to the combined efforts of the Government, through its health agencies, private physicians, nurses, etc., who have popularized the use of tikitiki extract, and to the education of the masses in proper infant feeding.

This situation did not last long. For the very next year, 1929, the incidence of the disease began to rise again and remained high for the next eleven years. Then from 1941 to the middle of 1946, another period of decline set in. In other words, the cycle from decline to decline seems to be 12-13 years. The first period of decline, lasting for three years, began in 1926; the next period of decline started in 1941 and continued up to the middle of 1946. From the other services in our Hospital (Medical and Obstetrical) and from other clinics in Manila, the same decline was noted. This period coincided with the Japanese occupation when food shortages became acute and death from starvation was an everyday affair. Generalized malnutrition, "hunger edema," xeropthalmia, keratomalacia, hyperkeratosis, and cancrum oris were frequently seen in our Out-Patient Clinics as well as in the wards. And yet infantile beriberi practically disappeared during this period.

e. Associated Diseases.—Infection plays an important though undetermined role in the etiology as well as the course and prognosis of infantile beriberi. Guerrero and Quintos(9) classified infantile beriberi into pure and mixed, the latter being associated with gastroenteritis, bronchitis or convulsions. In our cases, more than half were seen with some form of infection, either frank or subclinical, since fever, cough, or diarrhea was frequently present. The most commonly associated diseases were bronchopneumonia, bronchitis, and enteritis. These were often responsible for the flaring up of symptoms, and were often the cause of death in the cardialgic forms of beriberi.

f. Feeding.—All observers recognize breast-feeding as the condition "sine qua non" for the development of infantile beriberi. However, from time to time, cases occurring in bottle-fed infants are reported. Chapman(12) described 8 such cases in 1927. Since 1931, I have seen twelve cases among bottle-fed and forty-five among mixed-fed babies. In the last group, the information was usually obtained that the infants were nursed by their mothers for a period varying from one week to two months from birth. When the symptoms first became apparent, they were being artificially fed, and had been so for some time.

In view of all the above considerations, the conclusion seems inescapable that infantile beriberi is not simply a vitamin deficiency disease, and that other factors, besides lack of thiamin, are involved. Of course the most important factor seems to be faulty nutritional habits. The nursing mother of the stricken infant, when carefully questioned, usually revealed that she had been subsisting on a monotonous diet consisting of excessive quantities of polished rice, some fish, and little or no meats and vegetables. According to McCarrison(13) such a diet has at least four faults, namely, poverty in protein, excess of starch, deficiency of calcium, sodium and chloride, and lack of Vitamin A and B. Just exactly what role the secondary factors, enumerated above, play in bringing about the full-blown clinical picture of infantile beriberi cannot be determined at this time.

CLASSIFICATION

Various ways of classifying infantile beriberi have been proposed, based mostly on symptomatology. After studying our 968 cases, we believe that they can be divided into five groups.

1. *Pure Cardialgic, Fulminant or Pernicious Type.*

The onset is characteristically explosive. Vomiting after sucking may be the only prodromal symptom. The most important features in the clinical picture are as follows:

a. The baby is plump and apparently well nourished.

b. The baby's cry is a peculiar loud, piercing, and persistent scream repeated in paroxysms, apparently denoting severe suffering. This gradually gives way to a moaning or whining sound, as the child becomes exhausted.

c. The face is markedly pale, with a cyanotic tinge around the mouth.

d. The patient's body is stretched out, it may become stiff, and towards the end, convulsions may be noted.

e. The abdomen shows some rigidity, simulating flatulent colic.

f. Respiration is labored. In the agonal stage, the baby gasps for breath, the eyes staring upward.

g. Auscultation of the heart reveals accentuation of the second pulmonic.

h. Roentgenography shows an enlarged heart.

i. Therapeutic test with thiamin elicits a dramatic change in the clinical picture within one-half to six hours. On the other hand, cardio-tonics as caffeine, digitalis, and strophantin are ineffective.

The clinical picture derived from the above manifestations is almost characteristic. A baby around three months old, apparently in good health, nursed entirely by its mother, is abruptly seized with an attack of screaming. As he utters his loud piercing cry, his body stretches, the abdomen becomes hard, the pulse thready, the respiration labored, his face either deathly white or cyanotic, and an expression of profound terror or suffering grips his entire being. This state may last anywhere from one half to one hour. It disappears spontaneously, only to recur with increasing severity and frequency until death supervenes, or the specific treatment is promptly administered.

The Aphonic Type

Unlike the first group, this is characterized by an insidious onset and a long duration. It is much less serious than the cardialgic form. The outstanding feature is the dysphonia in some cases and complete aphonia in others. These cases usually begin with a slight fever and cough, or choking, and for this reason, are often mistaken for upper respiratory tract infection. As in the first, pallor of the face, with cyanosis around the mouth, is also seen. Restlessness, paroxysmal polypnea, oliguria and edema are often noted.

The aphonia may be severe. We saw a case in which restoration of the voice did not come until after some months, although all other symptoms had long since disappeared. When well developed, it also gives an unforgettable picture. As in the cardialgic type, the baby seems to be crying, but because of the loss of voice, no sound comes from him, and only his grimaces and twitching of his face offer evidence of the sufferings he is undergoing.

The laryngological examination is revealing. Alcantara and Ocampo(15) reported their findings in 37 cases in 1939:

"The five infants with acute cardialgic beriberi in whom impairment of voice was slight showed only congestion of the vocal cords, motility being normal. All the rest had impairment of motion. The right vocal cord was paretic or could not move to the median line in 4 cases, stayed immobile in the middle in 9 cases and assumed a cadaveric position in 3 cases. The left vocal cord was paretic in 5, completely immobile in the median line in 9 and cadaveric in 1 case. The vocal cords were bilaterally affected in 3 cases. In some cases, the paretic vocal cord appeared at a lower level than normal." This group showed the poorest response to thiamin.

The Pseudomeningitic Type

This type, first described by Albert in 1917, presents a distinct picture from the first two types. It is also less common, and is more often observed in older infants, between 6 and 12 months of age.

The typical picture is that of a well-nourished baby, breast-fed, who gradually becomes peaceful and quiet, as if he has forgotten how to cry or smile. He wears a languid and indifferent look, his eyelids are but half open. Sometimes, in addition to the ptosis of the upper lids, there is strabismus or nystagmus, suggesting encephalitis or tuberculous meningtis. There is, however, generally no nuchal rigidity. There may be spasmodic contraction of the facial muscles or choreic movements of the arms and hands. Sometimes there may be convulsions so severe as to require lumbar puncture. Vomiting and moderate constipation are noted. The temperature may rise to 38°C.

In 1934-1935, when we had an unusual number of these cases, the junior author made an observation which we thought might be useful in the differential diagnosis. We noticed that these cases of pseudomeningitic beriberi, in spite of their lethargic appearance, responded normally to certain stimuli. When a toy or any object was presented to one of these patients, in spite of his apparent stupor, he would reach out an unsteady arm to get the object offered to him. A similar stimulus given to patients with encephalitis or meningitis brought no response.

Often these cases are erroneously diagnosed as meningitis and progress to a fatal termination. Such errors are all the more tragic

because of the fact that these cases respond promptly to large doses of thiamin.

The Mixed Type

In this group are included those patients who show a combination of symptoms of the first three groups. Thus we have the (a) cardialgic-aphonic type (b) the cardialgic-pseudomeningitic, and (c) the aphonic-pseudomeningitic.

The cases that fall into any of the four groups described above are not hard to diagnose. There are, however, certain cases that present only a part of the symptom complex and thereby become major diagnostic problems. In some, the gastro-intestinal symptoms are most prominent, in others cyanosis. The diagnosis is often made by inference and the response to specific therapy. This group may probably be labelled the attenuated form or the "formes frustes."

THE PROBLEM OF THERAPY

Chamberlain and Vedder reported in 1912([18]) that an extract of rice polishings (tiki-tiki) was effective in beriberi. Since then confirmation has come from all parts of the world. The reduction of infant mortality in the Philippines from 65% to about 20% is considered to have been largely due to the extensive use of tiki-tiki extract. Its prophylactic and curative effects can no longer be doubted. In spite of this, however, we find that only about 31% of our cases were cured; the rest died or were discharged partly improved. We offer the following tentative explanation:

1. Those recovered were pure uncomplicated cases of the cardialgic and pseudomeningitic types who were brought in for early treatment.

2. Those that died were brought in too late or had some complicating acute infection.

3. Those that showed partial improvement only, without complete recovery, were mostly cases of the aphonic type. Although the symptoms of restlessness, pallor, edema, oliguria, etc. disappeared rapidly with the specific treatment, aphonia or dysphonia persisted for months. In this case, thiamin apparently had no effect on the paretic vocal cords.

These questions therefore come up: Is aphonic infantile beriberi true beriberi or something else? If it were true beriberi, why is

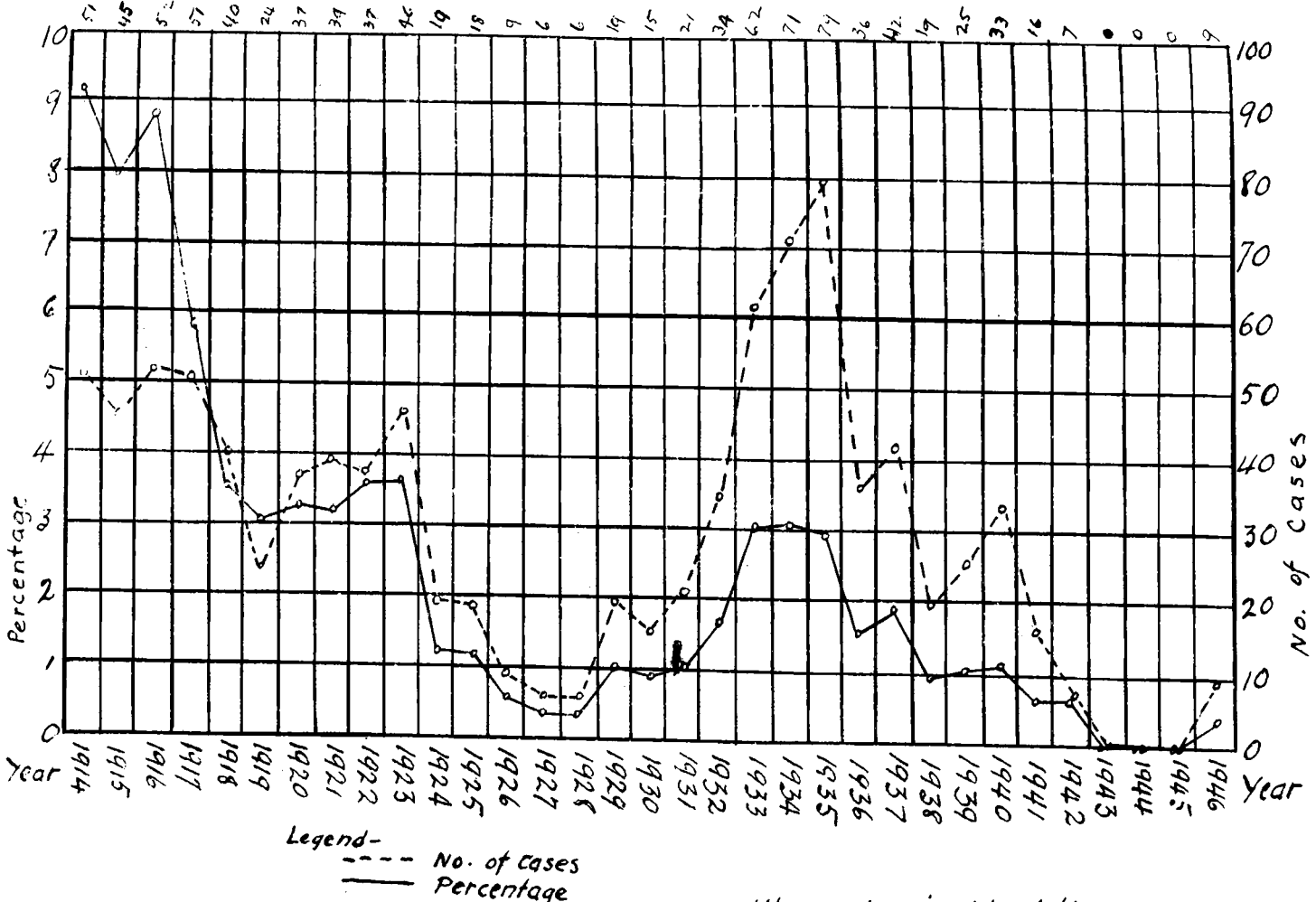


Fig. 1. Cases and Percentage of Infantile Beriberi with relation to Total Admissions

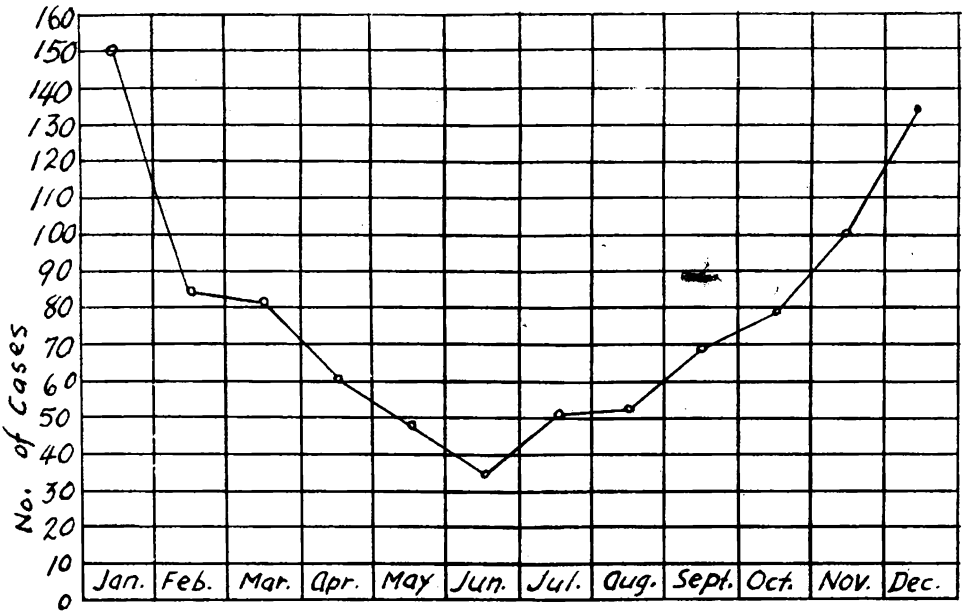


Fig. 2. Monthly Distribution of Inf. Beriberi

thiamin ineffective? Does it represent one of the irreversible stages of polyneuritis?

Remembering that most of the symptoms associated with the aphonia were quickly and favorably influenced by thiamin, one is inclined to consider that this type is a true beriberi.

Doubt about thiamin being an "antineuritic vitamin" has been expressed in various quarters. Thus it is well known that in adult beriberi it is ineffective, except for the relief of cardio-vascular manifestations. Aring and Spies(20) and Ming(21) believe that the initial prompt improvement with thiamin in cases of nutritional deficiency is humoral in nature. Peters(22) thinks that it acts as a catalyst in the carbohydrate metabolism of the nerve cell and of the heart muscle, so that its action in beriberi subjects is concerned with the reestablishment of the normal metabolism of the carbohydrate in the tissue. Walse(23) accordingly says: "To speak of Vitamin B_1 as 'antineuritic' is wholly erroneous except in relation to cases (human and avian) receiving a high carbohydrate diet." Meiklejohn(24) makes the following conclusions:

1. Thiamin is capable of curing a specific metabolic disturbance in the nervous system in animals. This disturbance has been incorrectly referred to as "polyneuritis" by many authors.

2. There is yet no clear experimental evidence showing that true anatomic polyneuritis in animals is curable by thiamin.

Thiamin deficiency in animals disturbs not only the normal metabolism of the nervous tissue, but also, and in a similar manner, the metabolism of the kidney, heart and probably other organs. It so happens that the metabolic disturbance of the nervous tissue manifests itself externally in a dramatic manner, while a similar disorder proceeding in the kidney and elsewhere produces no such obvious effects. This has given rise to the erroneous belief that thiamin has a specific effect on the nervous tissue. It is probably safe to say that thiamin is necessary for the normal metabolism of almost all tissues. It would seem that so far as the evidence from laboratory experiments is concerned, there is really no great justification for referring to thiamin as the "antineuritic vitamin."

SUMMARY

1. A study based on 968 cases of infantile beriberi seen in the Pediatrics wards of the Philippine General Hospital is presented.

2. The disease seems to have definite phases of intensity and decline, following a cycle of 12-13 years.

3. Seasonal variations, age, sex, etc. are definite secondary factors in the etiology.

4. The disease, as usually observed in our wards, usually occurs in association with upper respiratory tract or gastro-intestinal infections.

5. Classification of the disease, based on symptomatology, is presented.

6. Evidence that the condition is not due solely to vitamin B_1 deficiency is given.

REFERENCES

(1) FRASER, H. and STANTON, A. T.: Collected Papers on Beriberi. London. John Bale, Sons & Danielsson, Ltd. (1924).

(2) ALBERT, JOSE and OCAMPO, A. N.: Is Infantile Beriberi Disappearing? Jour. P. I. Med. Assoc., Vol. 8, No. 5, 221 224, May, 1928.

(4) Dr. MANAS, Puericulture Center of Makati, Rizal, and Dr. A. REGALA, Oroquieta Maternity and Children Hospital, Manila: Personal communications.

(5) OCAMPO, A. N., and HERRERA, N. A.: Avitaminosis A with Special Reference to Cutaneous Manifestations. Paper read at the Conference on Medical Sciences, Dec. 20, 21, 22, 1943, Manila.

(6) GAN, TOMAS M., MATIAS, M. Y. CRUZ, A. H., and SAN JUAN, JR.: Wartime Hypovitaminosis A: A Preliminary Report. Paper read at the Conference on Medical Sciences, Manila, Dec. 20, 21, 22, 1943.

(7) ALBERT, JOSE and PECACHE, L. V.: Incidence of Cancrum Oris since the Outbreak of the Greater East Asia War. Ibid.

(9) GUERRERO, M. and QUINTOS, J.: El Beriberi en los Niños de Pecho. Manila (1910).

(12) CHAPMAN, LELANDS S.: Infantile Beriberi in Panama. A Preliminary Report. Proc. Med. Ass. Isthmian Canal Zone 14 (1921-1925), 37-48. Abstract in Trop. Dis. Bull. 25 (June, 1928) 432.

(13) McCARRISON, R.: The Relationship of Diet to the Physical Efficiency of Indian Races. The Practitioner, Jan., 1925.

(15) ALCANTARA, V. C. and DE OCAMPO, G.: The Larynx in Infantile Beriberi. Archiv. Otolaryngology, Vol. 30, 389-399, Sept., 1939.

(16) ALBERT, JOSES La Forma Pseudomeningitica del Beriberi Infantil. Rev. Fil. de Med. y Farm. Vol. XI, No. 1, Jan., 1920, p. 11.

(18) CHAMBERLAIN, W. P. and VEDDER, E. B.: The Cure of Infantile Beriberi by the Administration to the Infant of an Extract of Rice Polishing, and the Bearing Thereof on the Etiology of Beriberi. Bull. Manila Med. Soc., 6 No. 2 (1912) 26-29.

(19) Liga Nacional para la Proteccion de la Primera Infancia. (Fifth Report to the Legislature. Rev. Fil. de Med. y arm., 10 Mayo, 1919).

(20) ARING, C. ET AL.: Some Clinical Neurologic Aspects of Vitamin B Deficiencies. J.A.M.A., 113 No. 24: 2105-2109, Dec. 9, 1939.

(21) MINZ, BRUNO: Le role de la vitamin B_1 dans la regulation humoraie du System Nerveux. Presse Med., 46: 1406-1407, Sept. 21, 1938.

(22) PETERS, R. A.: Lancel, 1: 1161-1165, 1936.

(23) WALSHE, F. M.R.: Aetiology of Polyneuritis. Lancet, 1: 33-35, Jan. 11, 1941.

(24) MEIKLEJOHN, A. P.: Is Thiamin the Antineuritic Vitamin? N. Eng. J. Med., 223: 265-273, Ang. 22, 1940.

(25) CLENDENING, L.: A Letter to the Editor, J.A.M.A., 117: 1036, Sept. 20, 1941.

TABLE I.—*Percentage of Infantile Beriberi*

Year	No. of Cases of Infantile Beriberi	Total Admission	Percentage of Infantile Beriberi
1914	51	547	9.32
1915	45	577	7.79
1916	52	591	8.79
1917	51	866	5.89
1918	40	1127	3.55
1919	24	792	3.03
1920	37	1091	3.39
1922	39	1171	3.33
1921	37	1030	3.59
1923	46	1276	3.61
1924	19	1369	1.39
1925	18	1383	1.30
1926	9	1408	0.64
1927	6	1316	0.46
1928	6	1344	0.45
1929	19	1573	1.15
1930	15	1712	0.88
1931	21	1853	1.13
1932	34	1955	1.74
1933	62	2092	3.01
1934	71	2318	3.07
1935	79	2767	2.85
1936	36	2377	1.51
1937	42	2306	1.82
1938	19	1953	0.97
1939	25	2008	1.24
1940	33	2390	1.38
1941	16	2157	0.70
1942	7	928	0.75
1943	0	916	0
1944	0	907	0
		From July 16-Dec. 31	
1945	0	916	0
1946	9	2648	0.34

TABLE II.—*Monthly Distribution of Cases*

Year	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Total
1914	5	4	3	1	1	2	2	2	6	6	11	8	51
1915	4	3	1	3	6	2	1	1	4	5	6	9	45
1916	8	6	3	2	2	3	3	3	1	4	8	9	52
1917	4	2	3	11	2	3	3	2	3	7	4	7	51
1918	5	7	6	6	1	2		1	2	1	1	8	40
1919	7	1	1	1	2	1		2	1	2	1	5	24
1920	3	1			2	1	1	1	4	4	7	13	37
1921	14	1	6	4	2		1		1	2	3	5	39
1922	3	2	4	3	3		3	3	5	2	3	6	37
1923	7	1	10	2	3	1	4	5	2	3	3	5	46
1924	3	2	2	2	2	0	2	4	1			1	19
1925	1	4	3	2	1	1	1	1		2	1	1	18
1926			2		1	1			2	1	1	1	9
1927					1			1	1	1	1	1	6
1928	2					1			1		1	1	6
1929	3	1	1		1	1	2	2		3	1	4	19
1930	1	1			1		3	3	3	2		1	15
1931	1	2	1	3		2	3	1		1	1	6	21
1932	5	2	3		1	1	1	4	2	4	6	5	34
1933	8	9	10	5	4	3	3	5	5	1	6	3	62
1934	5	8	5	1		3	6	3	3	8	7	22	71
1935	27	11	7	6	4	2			3	6	8	5	79
1936	9	2	4	2	4	1	1	2	4	3	3	1	36
1937	8	7	3	3	1	3	2	1	4	1	7	2	42
1938	4	4	1	2	1	1	2			2		2	19
1939	2	1	2				3	2	4	3	7	1	25
1940	8	3		2	2	1	1	1	4	3	4	4	33
1941	3	1	2	1	1		2	2	1	2	1		16
1942							2	1	3	1			7
1943													0
1944													0
1945													0
1946		1			1				4	1	1	1	9
Total	150	86	84	62	49	37	52	53	74	81	103	137	968

TABLE III.—*Incidence of Infantile Beriberi by Age Group*

Year	0-1 Wk.	2-4 Wks.	1-3 Mos.	4-6 Mos.	7-12 Mos.	Total
1914	1	9	34	4	3	51
1915	1	9	22	8	5	45
1916		7	33	9	3	52
1917		4	33	8	6	51
1918		4	19	8	9	40
1919		7	7	3	7	24
1920		5	20	7	5	37
1921		5	17	9	8	39
1922		7	18	6	6	37
1923	2	8	26	9	1	46
1924		2	12	2	3	19
1925		3	13	1	1	18
1926			2	4	3	9
1927			4	2		6
1928			3	1	2	6
1929			10	6	3	19
1930			4	3	8	15
1931			10	2	9	21
1932			18	10	6	34
1933			34	15	13	62
1934			29	20	22	71
1935			31	23	25	79
1936			18	15	3	36
1937		1	20	14	7	42
1938			10	6	3	19
1939			12	5	8	25
1940			18	12	3	33
1941			9	4	3	16
1942			2	1	4	7
1943						0
1944						0
1945						0
1946			6	3		9
Total	4	71	494	220	179	
Percentage	0.41	7.33	51.03	22.72	18.80	

TABLE IV

	1931	1932	1933	1934	1935	1936	1937	1938	1939	1940	1941	1942	1943	1944	1945	1946	TOTAL
A. No. of Cases	21	34	62	71	79	36	42	19	25	33	16	7	0	0	0	9	454
B. Types:																	
1. Cardialgic	13	20	26	43	36	16	16	6	5	12	4	2	0	0	0	4	203
2. Aphonic	3	7	26	6	11	9	7	2	4	9	6	2				2	94
3. Pseudomeningitic	5	6	5	7	11	3	4	1	7	5	1						55
4. Mixed:																	
a. Card.-Aphon.			1	6	10	7	14	9	8	7	5	2				3	72
b. Card.-Pseudo				1	2												3
c. Aphon.-Pseudo.				2								1					3
5. Frustrated: (attenuated)																	
a. Gastro-intes.				3	1												4
b. Cyanotic		1	4	3	8	1	1	1	1								20
C. Sex:				41	48	20											
1. Male	11	19	35	30	31	16	30	10	12	18	9	6				5	264
2. Female	10	15	27				12	9	13	15	7	1				4	190
D. Feeding:																	
1. Breast alone	19	27	61	55	71	30	39	18	21	31	12	4				9	397
2. Bottle fed		2		1	1	2	1	1		1	1	2					12
3. Mixed fed	2	5	1	15	7	4	2		4	1	3	1					45
E. Outcome:																	
1. Recovered	8	10	16	21	23	12	14	6	9	10	5					7	141
2. Improved	3	7	28	16	23	10	19	10	11	20	9	4				1	161
3. Unimproved					3	1				2							6
4. Died	10	17	18	34	30	13	9	3	5	1	2	3				1	146

PRIMARY ATYPICAL PNEUMONIA
THE AVERAGE CLINICAL PICTURE BASED ON 101 CASES *

PERPETUO D. GUTIERREZ, M.D.
FRANCISCO F. TANGCO, B.S., M.D.
AND
DOMINICO RUIZ, M.D.
Department of Medicine, College of Medicine,
University of the Philippines and the
Philippine General Hospital

It is only recently that most of us have given acceptance to that clinical entity first alluded to by Allen(1) in an analysis of 50 cases at Fort Sam Houston in 1935, characterized by a benign course, few physical signs, and X-ray evidence of a localized inflammatory process in the lungs—a form of respiratory infection, to designate which, he used the term "acute pneumonitis"; and which in 1938, Reimann(2) reported as atypical pneumonia, citing eight cases occurring in Philadelphia.

In 1942, Sison and co-workers read a paper(3) on the first cases reported locally. There was a temporary interest in the subject, but the majority of clinicians either were not convinced or considered the disease an extremely rare condition.

However, reports continued to come in of cases diagnosed as "lobar pneumonia" but which varied from the typical picture in certain respects. The most important difference was the gross disparity between the condition of the patient and the detectable physical findings. The patients were highly febrile, looked toxic, had a distressing cough and yet the internist, seeking verification through a meticulous physical examination, would find only some impairment of resonance, a few scattered rales and very little else. Moreover, the course of the disease was much shorter than the typical lobar pneumonia and more often than not, defervescence would be by lysis instead of by crisis. The difference in response to sulfonamides and penicillin was also striking.

Another facet of the problem became evident during the military campaign for the liberation of Manila. Shortly after the arrival of the American forces, cases of "atypical lobar pneumonia" began

* Read at the Thirty-ninth Annual Meeting, Philippine Medical Association, May, 1946.

to be reported in increasing numbers. In the light of the apparent increased incidence of this disease abroad during World War II(5) and of its admittedly high incidence in the Army(6) two questions may well be asked. Did the local population become more susceptible to the infection as a result of the breakdown of peacetime health safeguards, physical suffering and nutritional difficulties? Or were the people infected by carriers among the U. S. armed forces?

In an effort to throw some light on the main problem, this report is presented. We have studied the records of the 234 cases with the diagnosis of "lobar pneumonia" admitted to the Philippine General Hospital from July, 1945 to March, 1946. Of these, 101 cases deviated so clearly from the usual picture of lobar pneumonia and conformed to another recognizable pattern that we felt justified in setting them up as a distinct group.

Smith(7) has well summarized the present status of the entity, and we quote, "The increasing prevalence of an atypical form of primary pneumonia has commanded more and more attention . . . Much of the literature on the subject is puzzling or contradictory. There is no 'single criterion—clinical or laboratory—which characterizes the syndrome.'(8) Diagnosis is arrived at by a process of eliminating similar diseases of known etiology and attempting to check the patient's signs and symptoms against those of groups of cases previously reported. Unfortunately the literature on the subject is still too fresh for the relative value of each sign and symptom to have been worked out. Yet from the present maze of apparent contradictions among reported groups of cases, there is emerging a clinical picture sufficiently clear cut to permit reasonable diagnosis and tentative classification." Furthermore, these "several published accounts of a comparatively benign but special variety of pneumonitis conform so nearly to one pattern that there is little room for doubt that the disease should be considered as an entity."(9)

Nomenclature: We shall refer to the disease in this paper as primary atypical pneumonia. The designation assigned by the Commission on Pneumonia of the U. S. Army which probably applies to the largest standardized group of cases to be reported is "Primary, Atypical Pneumonia, Etiology Unknown."(10) It has also been referred to in literature as "Acute Pneumonitis,"(8) "Acute Respiratory Tract Infection, Type A," "Acute Interstitial Pneumonitis,"(11) "Bronchopneumonia of Unknown Etiology, Variety X,"(9) "Current Bronchopneumonia of Unusual Character and Undetermined Etio-

logy," 'Virus-type Pneumonia," "Viral Pneumonia," "Virus Pneumonia," and several others. Campbell et al believes it more nearly correct to call it "Acute Bronchiolitis with Associated Atelectasis."(12)

Predisposing Factors: In our cases, exposure to cold or rain was the most commonly mentioned factor. Fatigue also frequently preceded the attack, especially if there was subsequent exposure. Upper respiratory catarrh in the form of the "common cold" has apparently served to predispose some patients to the disease. Malnutrition did not seem to play an important role, as seventy four per cent of the cases were fairly nourished patients, about twenty per cent poorly nourished, and about six per cent were well nourished. None were emaciated.

Etiology: The causative organism of primary atypical pneumonia has not been identified. Recent and continued studies of this condition makes one feel that primary atypical pneumonia may prove to be not a single disease entity, but rather a clinical syndrome with multiple etiology.(14) A number of etiological agents have been mentioned—a number of known viruses, particularly of the psittacosis group,(15) rickettsiae, fungi, a protozoan that produces toxoplasmosis,(16) Coccidioides immitis,(17) and certain bacteria. Atypical signs and symptoms of pneumonia have also been observed during the migration of some of the parasitic merozoites.(18) A streptococcus (identified as No. 344) has been isolated in 2 cases of primary atypical pneumonia which terminated fatally, and it was shown that in fifty-five of one hundred and one cases, the patients had an increased titer to this organism.

Any of the aforementioned agents could produce a similar clinical syndrome but they can be excluded with reasonable certainty in the great majority of cases.(5) On the other hand, it was demonstrated that bacteria free filtrates obtained from sputum and throat washings, presumably containing a virus, can induce primary atypical pneumonia in man.(19) The results of the mass of laboratory experiments directed at isolating the etiologic agent leads only to the conclusion that primary atypical pneumonia is at least initiated, if not caused in its entirety, by a filter-passing agent, presumably a virus. The role of the bacterium in its causation is obscure.(20) It is possible that a single agent may be the cause of many or even most of the cases in a given outbreak, or in a single locality.(21)

Epidemiology: It is the consensus that primary atypical pneumonia occurs in epidemics. "Moist speakers" or impolite coughers,

birds and animals have been named as sources or transmitters of infections.

A common viral agent may have caused involvement of as high as fifty per cent of the population of some communities.(22) It seems that not all persons exposed to the disease acquire it. Persons with mild types of primary atypical pneumonia gave rise to the severer type in others, and vice versa. Children and infants are thought to be more susceptible than adults, although the higher incidence is among young adults. All ages, however, may be affected.(13)

In our series, one half of the total number of cases was found at ages from sixteen to twenty-five years, and the remaining half distributed among the later years, the number of cases decreasing as the age increased.

Both sexes are affected, but in our series, males were affected about four times as often as females.

Persons who engage in trades which require heavy manual exertion are apparently more susceptible. Fifty-nine of our cases were in the laboring class.

There is a distinct seasonal variation. In America it is most common in the fall and early winter, the incidence being highest during cold, damp, changeable weather, without any relation to the incidence of influenza or the common cold.(13) Our figures show the greatest distribution during those months of sudden changes of temperature, in December and during the transition to the hot months, in February. (Fig. 2). We have at the time of this report no figures for the months not included in this series (April to June).

Outbreaks are fairly common in crowded areas as in armies, schools, orphanages and jails.(13)

Morbid Anatomy: Since our series did not include a single case with a fatal termination, we cannot give a first-hand description of the pathological findings, and we have to depend entirely on what has been mentioned in the literature.

Grossly, the lungs resemble an acute miliary granulomatous process.(23) It is crepitant with isolated areas of pink or gray consolidation that vary in size.(24) Atelectasis and emphysema in other parts may be seen.(13) There may be hemorrhagic areas.(25) Infarcts have been reported.(9)

Microscopically, the fundamental pulmonic lesion is an acute interstitial pneumonitis.(23) Small bronchi, bronchioles and alveoli may be filled by frank pus(26) or a thick exudate of mucus, desqua-

mated cells, monocytes and a few neutrophils and eosinophils. Necrosis and ulceration of the epithelium may be found in the bronchi and bronchioles, with cellular debris filling the lumen.(25) The mucosa of the bronchi is inflamed and congested and bleeds readily.(27) Alveolar tissue is edematous, thickened and infiltrated primarily by monocytes.(24) This mononuclear alveolar exudate is peculiar to the disease.(9) A hyaline-like lining may be seen in the alveoli.(24) There may be metaplasia of the alveoli.(28)

Thrombosis and necrosis of the blood vessels with periarteritic changes may be seen.(24)(29)

Inclusion bodies in the epithelial cells have been seen and described.(25) This, however, is not specific for viral infections, for they may be due to Haemophilus pertussis, pasteurella tularense, toxins, irritative chemicals, or the protozoa of toxoplasmosis.(30)

Lymphangiectasis was invariably found.(26) Bacterial stains of lung sections uniformly failed to reveal micro-organisms in affected alveolar walls, alveolar lumens, peribronchial tissues, lung septa or bronchiolar wall.

Other pathological changes that have been noted were hemorrhage of the adrenal glands;(25) acute splenic tumor;(8) acute follicular splenitis with necrosis of enlarged malpighian corpuscles of the spleen;(13) focal necrosis of the liver;(24) edema of the meninges; congestion of vessels and small focal hemorrhages;(26) mesenteric lymphadenitis;(22) and hyaline necrosis of the diaphragmatic muscles.(31)

Symptomatology: In our patients, cough and fever were the most common presenting complaints. A great number of the patients consulted the physician because of severe headache which apparently did not respond to patent "cures." Chest and/or back pains or generalized body pains were also fairly common complaints. A few patients apparently sought the help of the physician because of persistent fever which had lasted for over two or three weeks.

The incubation period varies from five(32) to twenty-six days.(33) Reimann mentions, however, that it may be as short as one to two days. Fourteen to twenty-one days, or more precisely, seventeen to nineteen days, is believed to be the average duration.(34).

The onset was insidious in thirty-six of our cases and sudden in the remaining sixty-five. Literature is conflicting on this. Some claim an insidious onset as the rule,(14) allowing only twenty-five to thirty-three per cent for cases with sudden onset, while others,

like Daniels, reported a sudden onset in all his cases.(13) Page and Title, however, who gave an eighty-three per cent incidence of gradual onset would consider a lapse of two or more days between the onset of initial symptoms and the patient's hospitalization as gradual.

Fever was the most common symptom in our cases. In eighty-eight cases fever occurred early, as a rule on the first day. In eight it was a late symptom, occurring on the fourth day or even later. In five patients, fever was not noted during the whole course of the disease.

All kinds of temperature curves were observed. It may be high or only moderately so, ranging from thirty-eight to 40°C., or it may be only low-grade throughout. In forty-two cases, the temperature tended to be more or less continuous with only slight fluctuations. In twenty-nine patients it was of a remittent or "swinging" character.(13). In eighteen cases the fever was distinctly intermittent, going down to normal in certain hours of the day only to rise up in a few hours or so. A "dromedary" type of temperature was observed in seven cases, where there was fever for one or several days, normal temperature for two days or so, then another rise with exacerbation of symptoms. Lusk and Lewis' cases as well as some of Adams' presented such a "biphasic curve." In eighteen cases in the series there was a slight rise (up to 37.5°C.) that was observed after the defervescence and the patient had been afebrile and apparently symptom-free for some time.

The fever lasted from two to twenty-six days, the majority (seventy-five cases), being from four to twelve days. It may, however, last for forty-three days or even longer.(2)

Defervescence was by lysis in eighty-four cases, and by crisis in fifteen, the other two cases went home, against advice, still running a temperature.

Cough was the next most common symptom, occurring in eighty-seven of our cases. In only ten of these was cough noticed rather late in the course. Many times it came in paroxysms and tended to be more distressing or disturbing at night. It was usually dry at first but productive later on. Expectoration was mucoid or muco-purulent, whitish, yellowish, or greenish in fifty-nine cases. A brownish color, giving a rusty appearance, was seen in only five cases during the first three days of illness and, after this time, in five other cases. Six cases noticed blood streaks in the sputum instead.

Headache, an important early symptom(7), was rather common and many a time a very early and most disturbing symptom. It usually affected the entire head although some localized it at the frontal or temporal regions. It has been variously described as throbbing, crushing, or tightening, or just a dull aching pain. It was frequently so severe as to impair sleep and appetite and cause restlessness, and may not be relieved by the ordinary analgesics.

Chest and back pain were noticed at the onset in over half of the cases, although in others (twelve cases) it appeared rather late.

In about a third of the cases, the illness was ushered in by chilly sensations and in twenty-three others by actual shaking chills. Some had recurrent attacks of chills, while in others there was only one attack, appearing rather late in the course of the illness.

Only about a fifth of the cases actually complained of dyspnea or of chest oppression, and in nine cases only later in the course of the disease.

Other early symptoms observed were epigastic or generalized abdominal pain, tympanism, general body aches, joint, bone, or muscle pains, nausea and vomiting, anorexia, impairment of sleep, epistaxis, and profuse perspiration. Six patients had jaundice, ranging from a faint icteric tinge of the sclera to considerable yellowing of the skin. Bowel disturbances were present in some, in the form of frequent bowel movements, while in others there was constipation. Urine tended to be highly colored. There was actual polyuria in two cases. Coryza was complained of in three instances, while dizziness was one of the most disturbing symptoms of two cases. One patient had maculo-papular eruptions early in the disease. Restlessness, semiconsciousness, psychosis, hoarseness, slurring of speech, and laryngitis were among the rarer symptoms. Hebetude and body weakness were more common in the later stages. (Table 2).

The pulse rate in our cases increased more or less in proportion to the rise of temperature, a rate of 110 to 120 per minute being frequent, and rates as high as 150 having been observed. Bradycardia was not noticed.

The respiratory rate was only slightly increased, rates exceeding thirty per minute being rather infrequent.

Herpes labiales has not been observed.

Physical Findings: Physical findings initially were often confusing.(35) A patient may be bright and relatively comfortable only to show abundant physical findings, and another may look acutely ill and reveal little on physical examination.

The face was flushed in a few cases. Conjunctival injection was marked in about ten per cent of cases. In those cases where there was apparently either a toxic hepatitis or a concomittant hepatic involvement, scleral icterus was noted. Dilatation of the alae nasae on inspiration, as Campbell emphasizes, was conspicuous by its infrequency.(12) A dirty, furred tongue was rather frequent.

Slight rigidity of the neck was observed in one case.

Examination of the heart was essentially negative. In two cases, however, there was a functional murmur in the mitral area and in one case there was an apparent slight increase in the area of cardiac dullness. Accentuation of the second pulmonic sound was not infrequently observed.

The lung findings are very interesting. It is characterized by the great disparity between the complaints, the physical findings and the roentgenographic picture. In only 10 patients was there an appreciable limitation of the expansion of the affected side. In eighty-eight cases, the lesion was definitely patchy in nature, appearing apparently as if only a portion, or portions of the lobe was affected. Only impairment of resonance was appreciated in seventy-one cases, and dullness appeared only in twenty-one cases, while in the remaining nine, there were no percusory findings. Muscular hyper-irritability was found in two cases. Frequently, tactile fremitus was only slightly increased, in some it was decreased, and in a number there was no appreciable change.

Decreased breath sounds were rather common and were apparently at some time or another the only finding in some cases. Harsh breath sounds were appreciated in some cases and were absent in others. Bronchial breathing, brochophony, and whispered pectoriloquy were elicited in only a few instances. Rales were, as a rule, scanty, and may be crepitant, subcrepitant, sonorous, or sibilant. (Table II).

Any lobe of either or both lungs may be affected but basal lesions are most common—seen in eighty-six patients in our series, of which left-sided lesions were found in thirty-two cases, thirty-six cases with right-sided affection, and bilateral basal lesions in eighteen (Table IV).

Abdominal tenderness at one region or another was almost invariably elicited in those cases where abdominal pain was a symptom. In three cases, there was even some degree of rigidity.

Splenomegaly was appreciated in four cases but in all of these there was a very strong history of malarial infestation at one time

or another. Slight enlargement of the liver was observed in three cases without any attendant splenomegaly nor any history of malaria, and in one case where there was splenomegaly and malarial history.

Laboratory Examinations: There seems to be a tendency to slight anemia, over fifty per cent of our cases having counts of three to four million red blood cells per cu. mm. About twenty per cent had more or less normal counts. The hemoglobin content was on the average seventy to seventy-five per cent.

In the early part of the disease, half of our cases had counts ranging from 12,000 to 18,000. The greater bulk of the remainder had higher values, the highest count in 2 cases being over 35,000. There were, however, counts in the early stage below 7,000. There was a polynucleosis of from 76 to 95 per cent. Inadequate staining facilities, however, prevented us from ascertaining how much of this figure is made up of eosinophils, which may constitute a considerable percentage.(36) In the later part of the disease, there was observed, as a rule, a distinct and many times abrupt fall of the count, about two-thirds of the examined patients giving a count of from 7,000 to 13,000, of which seventy-one to eighty-five per cent were polymorphonuclears. The lowest count observed in the later stages was 4,200 and the highest 20,000. One case gave a count of 23,000, and another 35,000.

Urinalysis was essentially negative. In a few cases, there were traces of albumin, occasional hyaline casts, rare to few red blood cells, rare to few pus cells; in two cases there were abundant pus cells. Urinary findings were usually present during the highly febrile period, and disappeared soon after the drop of the temperature.

In those with suspected or manifest jaundice, elevation of serum bilirubin values was observed, Bilirubin I going as high as 0.641 mgm./cc. and Bilirubin II as high as 4.983. These values gradually went down to normal, pari passu with improvement of the case.

In all those cases where sputum examination was done, pneumococci were not identified. The mere presence of pneumococci in the sputum, however, does not necessarily rule out atypical pneumonia. There are reports of the isolation of the pneumococci in the sputum. These are not considered the causative organism since they were of the higher types.(12)

Roentgenological Findings: Inadequacy of supplies did not allow us to have the desired X-ray studies. Nonetheless, the examinations performed yielded very interesting and gratifying results.

The findings were very variable. We had cases where there was diminished transradiancy or diminished aeration of the affected side. This may appear more or less homogeneously over the whole lobe or field or may be in patches. Other findings were marked diffuse perihilar shadows; prominence of lung markings; cottony shadows which may appear like bronchopneumonic patches or congestion. A mottled appearance may also be observed. The shadow may also resemble lobar pneumonia but, as Green and Eldridge note, without obscuring the vascular and osseous markings.(44)

Complications: Complications of primary atypical pneumonia are apparently uncommon.

In our series we had a few. In six cases with scleral icterus and increased blood bilirubin values, there must have been at least a toxic hepatitis. Whether this is due, or not, to a primary hepatic involvement by the same etiologic agent, we are not in a position to determine. Suffice it to mention that in these cases the jaundice and the serum bilirubin values diminished and returned to normal as the patients improved.

Diaphragmatic pleuritis was observed in eight cases with an audible rub and referred abdominal pain. Meningismus was seen in two cases with negative spinal fluid findings. There was toxic psychosis in one patient who had complete abatement of symptoms with the recovery from the respiratory condition.

Evidence of pleural fluid was detected on the twelfth day in one case which on tapping yielded a thin sero-sanguinous exudate with four per cent albumin, 6,250 cells per cu. mm., with 66 lymphocytes, 21 polynuclears, 1 eosinophil, 6 macrophages, and 6 mesothelial cells. It was negative for any micro-organisms. This patient had an uneventful recovery and did not require a second tapping.

Prognosis: In the absence of any serious complication, concurrent or superimposed, prognosis is generally good. Reports of deaths, however, may be met in literature. The mortality rate in an Army camp with 1,862 cases was reported at 0.26 per cent(35) while among civilians, the rate is estimated at 2.4 per cent.(31)

Of the one hundred and one cases in the series, seventy were discharged "recovered," symptom-free and clear of any physical findings; twenty-nine were discharged "improved"—patients who were afebrile for sometime, completely symptom-free and with a normal blood picture, but still exhibiting some pulmonary physical findings, which may be in the form of persistent impairment of resonance,

bronchial breath sounds, or some moist rales; and two discharged against advice, "unimproved"—still running a temperature and with signs and symptoms.

The period of confinement ranged from three to forty days, the greater number staying in the wards for from one to two weeks.

Summary: The extremes and variations in the clinical and laboratory data of one hundred and one cases of primary atypical pneumonia were presented and compared with those reported in the literature.

REFERENCES

(1) ALLEN, W. H., "Acute Pneumonitis," Annals of Int. Med.: 10: 441, 1936.

(2) REIMANN, HOBART A., "An Acute Infection of the Respiratory Tract with Atypical Pneumonia," J.A.M.A., 111: 2377 (Dec. 24) 1938.

(3) SISON, A. B. M., JIMENEZ, V. B., and HERRERA, F., JR., "Virus Pneumonia," submitted for publication in the Jour. P.I.M.A. in Oct., 1942.

(4) IDSTROM, L. G. and ROSENBERG, B., "Primary Atypical Pneumonia," Bull. U. S. Army Med. Dept., No. 81 (Oct.) 1944.

(5) Commission on Acute Respiratory Diseases, "Atypical Pneumonia." Am. Jour. Med. Sciences, 209; 55 (Jan.) 1945.

(6) SMITH, RICHARD H., "Primary Atypical Pneumonia, Etiology Unknown," The Average Clinical Picture Based on Thirty-seven Original Cases," Annals Int. Med., 20: 890 (June) 1944.

(7) DINGLE, J. H. and FINLAND, M., "Primary Atypical Pneumonia of Unknown Etiology," New England. Jour. Med., 227: 378-385 (Sept. 3) 1942 Quoted by Smith, R. H. (5).

(8) LONGCOPE, W. T., "Bronchopneumonia of Unknown Etiology (Variety X)," A Report of Thirty-two Cases with Two Deaths," Bull. Johns Hopkins Hosp., 67: 268-305 (Oct.) 1940. Quoted by Schmitz (13).

(9) Official Statements, "Primary Atypical Pneumonia, Etiology Unknown," War Medicine, 2: 330-333 (1942).

(10) SMILE, D. R., SHOWACRE, E. E., LEE, W. F., and FERRIS, H. W., "Acute Interstitial Pneumonitis, A New Disease Entity," J.A.M.A., 112: 1901-1904 (May 13) 1939.

(11) CAMPBELL, T. A., STRONG, P. S., GRIER III, G. S., and LUTZ, R. J., "Primary Atypical Pneumonia; A Report of Two Hundred Cases at Fort Eustis, Virginia, J.A.M.A., 122: 723 (July 10) 1943.

(12) SCHMITZ, R. C., "Primary Atypical Pneumonia of Unknown Cause," Arch. Int. Med., 75: 222 (April) 1945.

(13) PAGE, S. G., JR., and TITLE, C. R., "Primary Atypical Pneumonia," Virginia Medical Monthly, 71: 305 (June) 1944.

(14) LEVINSON, D. C., GIBBS, J., and BEARDWOOD, J. T., JR., "Ornithosis as a Cause of Sporadic Atypical Pneumonia," J.A.M.A., 126: 1079 (Dec. 23) 1944.

(15) PINKERTON, H. and HENDERSON, R. G., "Adult Toxoplasmosis," J.A.M.A., 116: 807 (Mar. 1) 1941.

(16) GOLDSTEIN, D. and McDONALD, J. B., "Primary Pulmonary Coccidioidomycosis," J.A.M.A., 124: 557 (Feb. 26) 1944.

(17) KELLER, A. E., "The Clinical Manifestations, Treatment and Prevention of Ascaris," J. Tennessee M. A., 25: 346 (Sept.) 1932. Quoted by Schmitz(13)

(18) Commission on Acute Respiratory Diseases, Army Epidemiological Board, "Transmission of Primary Atypical Pneumonia to Human Volumeers": J.A.M.A., 127: 146 (Jan. 20) 1945.

(19) Commission on Acute Respiratory Disease, Army Epidemiological Board, "Present Status of Etiology of Primary Atypical Pneumonia," Bull. U. S. Army Med. Dept., 4: 494 (November) 1945.

(20) Editorial, "The Virus of the So-called 'Virus Pneumonias'," New Eng., J. of Med., 228: 729 (June 3) 1943.

(21) Editorial, "'Acute Pneumonitis' Virus," J.A.M.A., 116: 1222 (Mar. 22) 1941.

(22) Editorial, "Primary Atypical Pneumonia," J.A.M.A., 126: 18 (Dec. 30) 1944.

(23) KNEELAND, Y., JR., and SMETANA, H. F., "Current Bronchopneumonia of Unusual Character and Undetermined Etiology," Bull. Johns Hopkins Hosp., 67: 229 (Oct.) 1940. Quoted by Schmitz(13)

(24) ADAMS, J. M., "Primary Virus Pneumonitis with Cytoplasmic Inclusion Bodies," J.A.M.A., 116: 925 (Mar. 8) 1941.

(25) GOLDEN, A., "Pathology of Atypical Pneumonia," Bull. U. S. Army Med. Dept., 81: 64 (Oct.) 1944.

(26) DINGLE, J. H., ABERNETHY, T. J., BADGER, G. F., BUDDINGH, G. J., FELLER, A. E., LANGMUIR, A. D., RUEGSEGGER, J. M., and WOOD, W. B., JR., "Primary Atypical Pneumonia, Etiology Unknown," Am. Jour. Hygiene, 39: 67 (Jan.) 1944. Quoted by Schmitz(13).

(27) LONGCOPE, W. T., "Atypical Virus Pneumonia," Bull. New Eng. M. Center, 4: 21 (Feb.) 1942. Quoted by Schmitz(13).

(28) THOMAS, H. M., JR., "The Role of Alpha Hemolytic Streptococcus in Pneumonia," Bull. Johns Hopkins Hospital, 72: 218 (April) 1943. Quoted by Schmitz(13).

(29) SABIN, A. B., "Toxoplasmic Encephalitis in Children," J.A.M.A., 116: 801 (Mar.) 1941.

(30) OWEN, C. A., "Primary Atypical Pneumonia: An Analysis of Seven Hundred and Thirty-eight Cases Occurring During 1942 at Scott Field, Ill.," Arch. Int. Med., 73: 217 (Mar.) 1944.

(31) KAMIN, H. N., "Virus Pneumonia," Illinois, Med. Jour., 83: 41 (Jan.) 1943.

(32) GOODRICH, B. E., and BRADFORD, H. A., "The Recognition of Virus Type Pneumonia," Am. J. M. Sc., 203: 163 (Aug.) 1942.

(33) a. DINGLE, J. H., ABERNETHY, T. J., BADGER, G. F., BUDDINGH, G. G., FELLER, A. E., LANGMUIR, A. D., RUESEGGER, J. M., and WOOD, W. B., "Primary Atypical Pneumonia, Etiology Unknown," War Medicine, 3: 223 (Mar.) 1943.

b. CAMPBELL, E. T., "Primary Atypical Pneumonia and Malaria," War Medicine, 3: 249 (Mar.) 1943.

(34) VAN RAVENSWAAY, A. C., ERICKSON, G. C., ROH, E. P., SIEKERSKI, J. M., POTTASH, R. R., and GUMBINER, B., "Clinical Aspects of Primary Atypical Pneumonia (A Study Based on 1,862 Cases Seen at Station Hospital, Jefferson Barracks, Missouri," J.A.M.A., 124: 1 (Jan. 1) 1944.

(35) CONTRATTO, A. W., "So-called 'Atypical Pneumonia' Among College Students," New Eng.J. Med., 229: 229 (Aug. 5) 1943.

(36) GUNDERSEN, S., "Primary Atypical Pneumonia of Unknown Etiology," New Eng., J. Med., 231: 697 (Nov. 23) 1944.

(37) HELWIG, F. C. and FREIS, E. D., "Cold Autohemagglutinins Following Atypical Pneumonia Producing the Clinical Picture of Acrocyanosis," J.A.M.A., 123: 626 (Nov. 6) 1943.

(38) EMERSON, K., JR., ET AL., "Chloride Metabolism and Plasma Amino Acid Levels in Primary Atypical Pneumonia," J. Clin. Investigation, 22: 695 (Sept.) 1943. Quoted by Schmitz(13).

(39) RAKE, G., EATON, M. D., and SHAFFER, M. F., "Similarities and Possible Relationships Among Viruses of Psittacosis, Meningipneumonia, and Lymphogramuloma Venereum," Proc. Soc. Exper. Biol. and Med., 48: 528 (Nov.) 1941. Quoted by Schmitz(13).

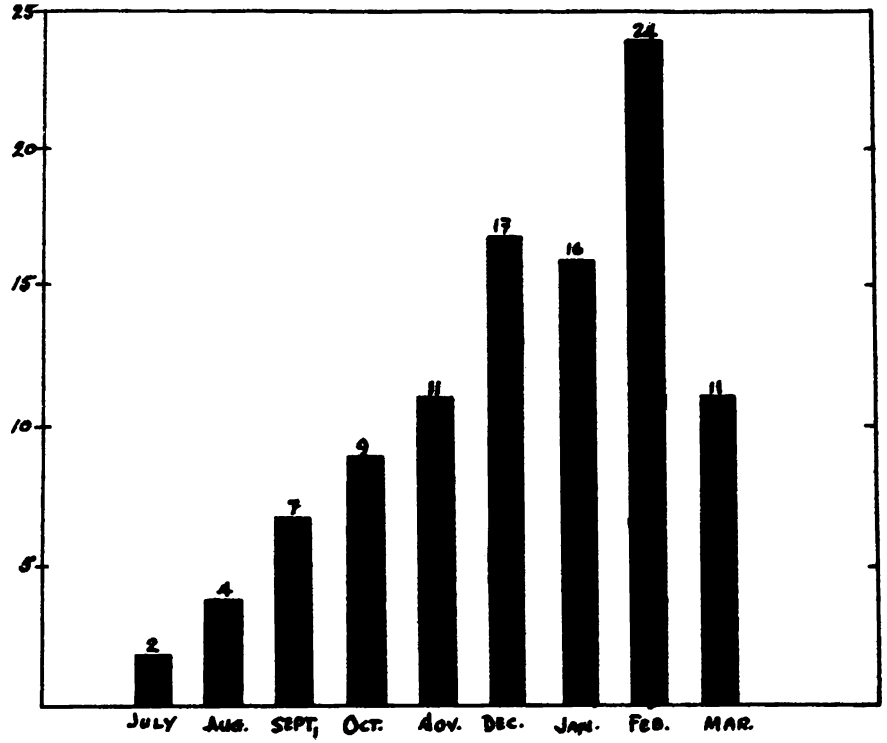
(40) GALLAGHER, J. R., "Bronchopneumonia in Adolescence," Yale J. Biol. and Med., 7: 23 (Oct.) 1934. Quoted by Schmitz(13).

(41) SMITH, J. H. "Virus Pneumonia by Contrast with Other Types," Virginia Medical Monthly, 70: 353 (July) 1943.

(42) OFFUTT V. D., "Diagnosis and Treatment of Primary Atypical Pneumonia," Virginia Medical Monthly, 71: 431 (Aug.) 1944.

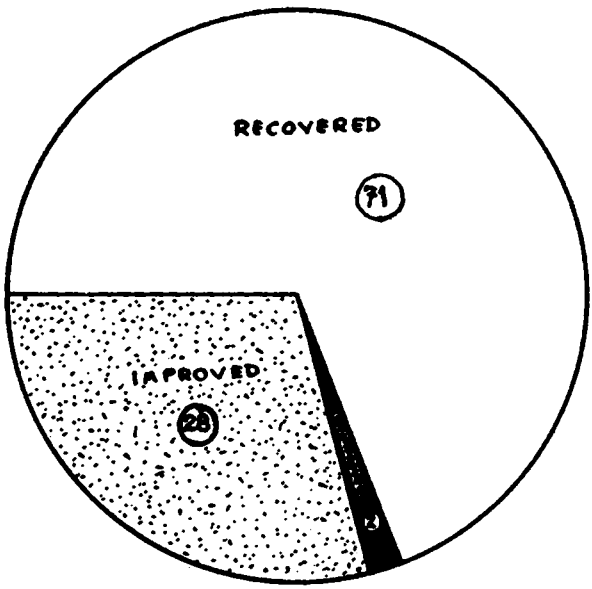
(43) GREEN, D. M. and ELDRIDGE, F. G., "Primary Atypical Pneumonia, Etiology Unknown," Military Surgeon, 91: 503 (Nov.) 1942.

(44) RACKER, E., ROSE, S. P., and TUMEN, A. O., "Pneumococcic Pneumonia Resembling Primary Atypical Pneumonia," Am. J. M. Sc., 209: 496 (Apr.) 1945.



Monthly Distribution of the 101 Cases of the Series

N.B. There were no figures obtained for April, May, and June



Condition on Discharge

TABLE I.—*Symptoms*

	First 3 days	*Later*
Fever	88	8
Cough	77	10
Chest pain	57	7
Mucoid expectoration	54	5
Back pain	44	5
Headache	40	3
Chilly sensations	31	
Chills	23	5
Dyspnea	21	9
Abdominal pain	11	4
Vomiting	10	4
Muscle pains	6	
Icteric sclerae	6	1
Bone pains	6	1
Anorexia	6	4
Rusty sputum	5	5
Blood streaks in sputum	4	2
Nausea	4	1
Coryza	3	
Epistaxis	3	
Dizziness	2	
Impaired Sleep	2	1
Body weakness	2	6
Chest oppression	2	
Epigastric pain	2	1
Frequent urination	1	1
Highly colored urine	1	2
Incoherence	1	1
Insomnia	1	1
Skin eruptions	1	
Psychosis	1	
Profuse perspiration	1	1
Semiconsciousness	1	1
Tympanism	1	
Restlessness	1	
Laryngitis	1	

TABLE II.—*Physical Examination*

Nutrition:	
Well nourished	6
Fairly nourished	75
Poorly nourished	21
Emaciated	0
Head:	
Flushed face	2
Dilatation of alae nasae	8
Coated tongue	32
Injected conjunctivae	8
Icteric sclerae	10
Neck:	
Slight rigidity	1
Chest:	
Heart:	
Murmur, soft blowing	2
Enlarged, 6th I. S.	1
Accentuated 2nd pulmonic sound	21
Lungs:	
Inspection:	
Limited mobility of affected part	10
Percussion:	
Muscular hyper-irritability	2
Dullness	21
Impaired resonance	71
Palpation:	
Tactile fremitus increased	69
Tactile fremitus normal	11
Tactile fremitus decreased	21
Auscultation:	
Breath sounds weak	68
Breath sounds absent	3
Breath sounds harsh	5
Bronchophony	4
Bronchial breath sounds	10
Whispered pectoriloquy	0
Rales:	
Crepitant, abundant	15
Crepitant, few	60
Subcrepitant, few	33
Subcrepitant, abundant	10
Sonorous	5
Sibilant	9
Rub	5
Abdomen:	
Splenomegaly	4
Hepatomegaly	4
Rigidity	3
Tenderness:	
Right hypochondrium, slight	4
Left hypochondrium	2
Right iliac	1
Left iliac	1
Epigastrium	1

TABLE III.—*Character of Fever*

	No. of cases
With slight rise after falling to normal	18
Continuous:	
High	22
Moderately high	20
Remittent:	
High	12
Moderately high	17
Intermittent:	
High	6
Moderately high	10
Low	2
Dromedary	7

Completely afebrile	5
With slight rise after falling to normal	18

TABLE IV.—*Lung Involvement*

	Patchy	*"Lobar"*
Right:		
Upper lobe	3	1
Middle lobe	5	0
Base	28	4
Upper, middle	0	0
Middle, base	4	2
Left:		
Upper lobe	2	0
Base	26	4
Both lobes	2	0
Bilateral:		
Upper lobes	1	0
Bases	17	1
Mixed	1	0
TOTAL	89	12

TABLE V.—*Differential Criteria*

	Pneumococcic	*Atypical*
Onset	Abrupt	Slow
Cyanosis and Dyspnea	Frequent	Rare
Herpes	Frequent	Rare
Pulse rate	Rapid	Normal or slightly accelerated
Respiratory rate	Accelerated	Normal
Physical signs	Impaired resonance	Very slight change in resonance
	Bronchial breathing	Rare bronchial breathing frequent rales
Sputum gross	Rusty	Greenish mucoid
Sputum microscopic and culture	Pneumococcic	No predominating organism
White blood cell count	High	Normal
X-ray finding	Dense consolidation	Stringy and mottled type density
Crisis:	Frequent	Rare
Response to Sulfonamides	Good	None

A STUDY ON IMMUNITY IN 587 POSITIVE SMALLPOX VACCINATION *

MANUEL Y. MATIAS and AMADEO H. CRUZ
The Institute of Hygiene
University of the Philippines

INTRODUCTION

In 1923 Kolmer laid down the conditions that must be satisfied in order that infection can take place. He propounded that infection can take place only when:

1. The organisms are sufficiently virulent.
2. When they invade the body by appropriate avenues and reach susceptible tissues.
3. When they are present in sufficient numbers.
4. When the host is generally susceptible to their action.
5. When the microorganisms are able to resist the defensive forces of the host through special agencies apart from their offensive forces.

Stallbrass (1931) and Lara (1937) simplifying the above requisites agree in that the basic fundamental principle of disease production is the presence of the following:

1. Causative microorganisms.
2. Susceptible individuals.
3. Appropriate means of conveying the aetiology to the susceptibles.

The latter calls them the primary factors of diseases causation and further adds that these may be affected by a multiplicity of secondary factors. On the basis of this postulate, smallpox cannot appear unless the smallpox virus, susceptible persons and appropriate avenue of bringing the smallpox virus to the susceptible are all present.

In smallpox, there seems to be as yet no practical means of demonstrating the etiological agent as we are here dealing with a virus difficult of demonstration.

---

* Read at the Thirty-seventh Annual Convention, Philippine Medical Association, March 8, 1940.

Smallpox vaccination which is primarily used for prophylactic purpose can, however, be utilized as a test of the presence or absence of immunity.

In any community where this disease is non-existent, the permanent absence of this disease can be maintained only by cutting the link connecting the primary factors of its causation so that these cannot successfully operate. The fight can be carried on at the "seed front"; it can be carried on at the "sower front." We all know, however, that it is most decisive when brought to the "soil front," that is by means of smallpox vaccination.

OBJECT OF THIS STUDY

The object of this investigation is to determine the number of susceptibles based on the type of local reaction in the group of persons examined, and incidentally to find out approximately how long a successful vaccination protects the individual. To make this study possible the authors took advantage of the annual smallpox vaccination in the public primary and elementary schools in the district of Paco. To augment our cases, we included the results of the vaccination of the third year medical students of the College of Medicine, University of the Philippines.

During the months of July, August and September of 1939, the authors, assisted by a staff of nurses, vaccinated all the first and seventh grade pupils of the public schools mentioned above. The multiple pressure method was employed. As part of the training of the third year medical students in preventive medicine, they were required to perform vaccination among themselves. Results of the vaccinations of this group were added to the total number investigated. Following the universally accepted classification of the different types of local reaction that may follow smallpox vaccination, we grouped the positive reactions into:

1. Primary reaction
2. Secondary reaction
3. Immediate reaction

A positive reaction is considered primary, secondary, or immediate on the basis of definite manifestations at the site of vaccination appearing in an orderly manner.

Primary reaction (synonyms: vaccinia, "take," reaction of non-immunity) which indicates absence of immunity in the person prior

to vaccination, is characterized by an incubation period of three days, followed successively by the papular, vesicular and pustular stages of three days duration each. The pustular stage becomes fully mature on the twelfth day and thereafter rapidly subsides, leaving a foveated scar.

The secondary reaction (synonyms: accelerated reaction, vacciniod reaction, reaction of partial immunity) indicates that the person reacting in this way still has a certain amount of immune bodies in his tissues and fluids. It is characterized by an incubation period of about thirty-six hours followed successively as in the primary reaction by the papular, the vesicular and the pustular stages, the latter becoming fully mature on the eighth day and thereafter dries up rapidly leaving a scar less uneven and shallower than that produced by the primary reaction.

The immediate reaction (synonyms: reaction of complete immunity, immune reaction) indicates that the person is still completely and absolutely immune to the disease. It exhibits only two stages, an incubation period of twenty-four hours and a papular stage of from three days to three weeks.

While the Bureau of Health (Circular No. 668 of the Director of the Bureau of Health, November 14, 1934) classifies the "immediate reaction" as a negative reaction for purposes of recording, in this paper it is considered a positive one.

Our negative reaction, therefore, includes only those showing no manifestation at all at the site vaccinated except for a slight transitory reddening caused by trauma which disappears in a few hours.

Since the negative results will not be of use in the present study we purposely left them out of consideration unless they become positive on a subsequent vaccination.

RESULTS AND DISCUSSION

Results of our study are consolidated in Tables I, II and III. Table I gives us a consolidated record of the results of all the 578 positive vaccinations arranged by types of local reaction, age group and sex distribution. Table II gives us the percentage distribution by types of reaction in each age group. Table III gives us the percentage distribution by age group and by types of reaction.

Of the 587 positive vaccinations that we recorded, only very few did not conform strictly to the sequence of changes described above. These we placed under the type to which they conform most closely.

It will be noted in Tables I and II that of a total of 289 falling within the age group 5-9 years, 184 or 63.67 per cent were without immunity when revaccination was performed, that 71 or 24.57 per cent were partially immune, and 34 or 11.76 per cent were still completely immune when they were vaccinated in the present series. Of the total 170, falling within the age group 10-14 years, 64 or 37.65 per cent were without immunity, 89 or 52.35 per cent were partially immune; and 17 or 10 per cent were still completely immune when vaccinated. Of the total 51 falling within the age group 15-19 years, 17 or 33.33 per cent were without immunity; 25 or 49.02 per cent were partially immune, and 9 or 17.65 per cent were still completely immune. Of the 75 falling within the age group 20-24 years, 11 or 15.06 per cent were without immunity on the day of vaccination, 34 or 46.58 per cent were still partially immune, and 28 or 38.36 per cent were still completely immune. Of the total 4 falling within the age group 25-29 years, 1 or 25 per cent was without immunity, another 1 or 25 per cent was partially immune, and 2 or 50 per cent were still completely immune at the time of the vaccination in this series.

In this paper we are particularly interested in the age group 5-9 years for obvious reasons, hence we shall limit our discussion to this as much as possible.

Sections 1051-1057 of the Revised Administrative Code (Act No. 2711), Act No. 3754 amending Section 1056 of Act. No. 2711 and Circular R-55 of the Director of Health (July 15, 1919) provide, among other things, for compulsory vaccination against smallpox during infancy and school age. Act No. 3573 (Approved November 26, 1929) provides compulsory inoculation with a prophylactic preparation of recognized efficiency. Circular T-52 of the Bureau of Health (June 17, 1921) provides for a systematic vaccination against smallpox among those who attend primary school for the first time and among those newly enrolled in the high school.

Many parents are aware of the benefits derived from vaccination so that infants who might escape the attention of the health officer on account of non-registration of their birth, are taken to

the health officer for immunization. There are parents who are ignorant of the law requiring compulsory birth registration but practically all know the protection conferred by vaccination, so they seek the health officer if the latter fails to look for them so that their infants could be vaccinated. In view of this and the routine house to house visit of the nurses of the Urban Health Demonstration Unit (whose field of activities embraces the whole district of Paco), it is very difficult for a birth to escape registration. Invariably every birth accounted for is followed by vaccination. It is worth mentioning here that it is the practice of the Bureau of Health and the Unit personnel to repeat the vaccination as many times as is necessary until a previously negative one becomes positive.

Since under the foregoing circumstances it would be very difficult for a child to escape vaccination during infancy, it is presumed that all of the 289 pupils within the age group 5-9 years were rendered immune when they were one year old or thereabout. Putting it in another way, the vaccination they had in this study is supposedly their second positive vaccination.

The observation that 184 out of the 289 gave the reaction of non-immunity indicates that 63.67 per cent have already lost their immunity. The number of susceptibles among the first graders is very high. If the proportion holds true in all other private and public schools in the Philippines, and if to them is added the number of susceptibles in other age groups we certainly have a very high percentage of susceptibles in our community. The population of the Philippines for age-groups 1-9 years as obtained from the Census of 1932 is 4,660,637. This will give us an idea of the number of susceptibles in this age group.

Circular No. Q-17 of the Director of Health (dated April 12, 1918) reads in part, "the spread of smallpox epidemic in Manila and in some provinces of the archipelago has shown that immunity conferred by the general vaccination in the years 1909 and 1910 is being lost." From the above, we can deduce that the length of immunity conferred upon the populace vaccinated in 1909 and 1910 was approximately seven years. The conclusion is based on a rough generalization.

Circular No. V-41 (May 31, 1923) provides for the revaccination of the entire population every seven years. This step agrees in every respect with the implied length of immunity deduced from the provision of Circular No. Q-17.

Circular No. 148-C (September 6, 1937) of the Director of Health reads in part, "the duration of the immunity conferred by anti-smallpox vaccination having been estimated in general to last 5 to 7 years, all concerned are hereby instructed, for the sake of economy and in order to avoid waste of time, to refrain from vaccinating children in the 1-4 years age group, if they exhibit scar from previous successful vaccination (previous positive)." While much has been said about the length of immunity conferred by smallpox vaccination all were based on studies made abroad and the subjects were not Filipinos.

The subject matter is something that is not new, but no studies conducted locally that we know of have been made among Filipinos. Small as our cases are, they give us information about Filipino subjects. We have stated somewhere before that it is very difficult for an infant in the District of Paco to escape vaccination. Because of this we presume that every one of the 289 5-9 years old first grade pupils vaccinated must be immune to the disease. This is of course taking things for granted. It would have been better if all the 289 5-9 years old pupils were vaccinated by us during infancy.

This paper is being presented in the form of a preliminary report. Whatever conclusion we deduce is tentative. Since the 289 5-9 years old pupils mentioned above gave the reaction of non-immunity, they must have lost the immunity we presumed they acquired during the first vaccination. Considering the midpoint of the age group as the mean age of the 289 pupils, it may be said that at 7½ years of age, 63.67 per cent of the school children examined have entirely lost their immunity against smallpox, since at this mean age 63.67 per cent gave the reaction of non-immunity. Just exactly at what age they lost their immunity, whether at the second year, at the third year, at the fourth year, at the fifth year, or at the sixth year of age is a question difficult to answer at the moment. Approximately, however, the duration of the immunity cannot be far from 7½ years. Experience has shown us that the usual age of the infants for vaccination is from four months to ten months. Now, if we consider the midpoint of 0-1 year as representing the average age when most infants were immune, then approximately the maximum length of immunity in the group of 289 school children is seven years (7-½ years minus 6 months).

Dunham gives 2-10 years as the duration of immunity in general following smallpox vaccination, less than two years and more than ten years in rare instances. Dearing and Rosenau showed in 1934 from Massachusettes statistics that one vaccination usually protects for at least twenty years. In the epidemic of 1924 in Detroit (Vaughan, et al., 1925), no one with a vaccination of less than five years duration contracted smallpox. Our experience, however, shows that there is need of more frequent revaccination.

SUMMARY AND COMMENT

1. As shown by the types of local reaction, there is a high percentage of susceptibles among the groups of people examined, particularly among school pupils within the age group 5-9 years, corresponding to those entering school for the first time. If the proportion hold true in all other private schools, we have a very great area of fertile soil for the development of smallpox. Our findings suggest earlier first revaccination in order to prevent or at least minimize the increase of susceptibles in those losing their immunity from the first vaccination.

2. Evidence indicates that the immunity obtained in the first vaccination lasts for seven years.

ACKNOWLEDGMENT

The authors wish to thank Dr. Jose Sian of the Bureau of Health for his kindness in lending to us the compiled circulars, memoranda, etc. about smallpox vaccination in the Philippines.

REFERENCES

1. DUNHAM, G. C.: 1938 — Military Preventive Medicine, 3rd. Ed., pp. 93-101, Pennsylvania: Army Medical Bulletin No. 3, Carlisle Barracks.
2. GRUBS, S. B.: 1923 — Vaccination Technique and Certification, An Experiment in Making Vaccine and Insurance Against Delay as Well as a Protection Against Disease. Public Health Reports, Reprint No. 869, Government Printing Office, Washington, pp. 2201-2206.
3. LARA, H.: The Law of Disease Incidence. Read in the 4th Philippine Science Convention, February, 1937.
4. Revised Administrative Code of 1917 (Act 2711) Sections 1051 — 1057.
5. STALLBRASS, C. O.: 1931 — The Principles of Epidemiology. New York. The MacMillan Co. pp. 76-123.

6. Smallpox and Vaccination. Monthly Bulletin of the Philippine Health Service, 12:15-16 (Jan.) 1932.
7. ROSENAU, M. J.: 1931 — Preventive Medicine and Hygiene, 5th. Ed. pp. 1-32, New York, D. Appleton & Co.
8. LOPEZ-RIZAL, L.: 1924 — Vaccination and Smallpox in the Philippine Islands, Transactions of the Fifth Biennial Congress of the Far Eastern Association of Tropical Medicine, edited by Dr. A. L. Hoops and Dr. J. W. Schaeff, pp. 538-562, London: Sons and Danielson, Ltd.
9. The Reactions Which Follow Smallpox Vaccination. Baltimore Health News, Baltimore Health Department, 13: 17-18 (April-June) 1936.
10. Act 3754 of the Philippine Legislature Amending Section 1056 of Act 2711, Approved November 26, 1929.
11. Act 3573 of the Philippine Legislature, Approved Nov. 26, 1929.
12. Circular No. Q-17 of the Director of Health dated April 2, 1918.
13. Circular No. R-55 of the Bureau of Health dated July 15, 1919.
14. Circular No. T-52 of the Director of Health dated June 17, 1921.
15. Circular No. U-41 of the Bureau of Health dated May 31, 1923.
16. Circular No. 668 of the Director of Health dated Nov. 14, 1934.
17. Circular No. 148-C dated September 6, 1937 of the Bureau of Health.

TABLE I.—*Types of Local Reaction by Age and Sex in 587 Positive Vaccinations*

Age Group	Primary Reaction			Secondary Reaction			Immediate Reaction			Total
	Male	*Female*	*Both Sexes*	*Male*	*Female*	*Both Sexes*	*Male*	*Female*	*Both Sexes*	
5-9	94	90	184	40	31	71	19	15	34	289
10-14	36	28	64	48	41	89	9	8	17	170
15-19	14	3	17	15	10	25	3	6	9	51
20-24	7	4	11	22	12	34	16	12	28	73
25-29	1	0	1	1	0	1	2	0	2	4
TOTAL:	152	125	277	126	94	220	49	41	90	587

TABLE II.—*Percentage Distribution of Types of Reaction in Each Age Group*

Age Group	Primary Reaction		Secondary Reaction		Immediate Reaction		Total	
	Number	*Per cent*	*Number*	*Per cent*	*Number*	*Per cent*	*Number*	*Per cent*
5-9	184	63.67	71	24.57	34	11.76	289	100
10-14	64	37.65	89	52.35	17	10.00	170	100
15-19	17	33.33	25	49.02	9	17.65	51	100
20-24	11	15.06	34	46.58	28	38.36	73	100
25-29	1	25.00	1	25.00	2	50 00	4	100
TOTAL:	277		220		90		587	
Percentage	47.19		37.48		15.33		100	

TABLE III.—*Percentage Distribution by Age Group in Each Type of Local Reaction*

Age Group	Primary Reaction		Secondary Reaction		Immediate Reaction		Total Number	Percentage
	Number	*Per cent*	*Number*	*Per cent*	*Number*	*Per cent*		
5-9	184	66.43	71	32.27	34	37.78	289	49.23
10 14	64	23.10	89	40.46	17	18.89	170	28.96
15-19	17	6.14	25	11.36	9	10.00	51	8.69
20-24	11	3.97	34	15.46	28	31.11	73	12.44
25-29	1	0.36	1	0.45	2	2.22	4	0.68
TOTAL:	277	100 00	220	100.00	90	100.00	587	100.00

PROTHROMBIN STUDIES AMONG FILIPINOS

R. J. NAVARRO, M.D., E. N. FERMIN, M.D., and R. SOLIS
College of Medicine, University of the Philippines and the Philippine General Hospital

The presently accepted theory of blood coagulation assumes that there are four elements concerned in this process—prothrombin, fibrinogen, calcium and thromboplastin.

Three important stages are assumed in the process of clotting:

1. The formation of thrombin from prothrombin. As soon as blood extravasates from a vessel, thromboplastin from the degenerated blood platelets and from the tissue juice acts on the prothrombin liberating thrombin in the presence of calcium.

2. The reaction of thrombin with fibrinogen to form fibrin which is the essential element of the clot.

3. Retraction of the clot.

To explain fully the hemorrhagic manifestations of diseases it will be necessary, therefore, to determine which element of blood coagulation is deficient. Fibrinogen and calcium can well be determined quantitatively by standard methods. Since thromboplastin is in great part derived from the blood platelets, a blood platelet count could serve as a basis for measuring thromboplastin activity. For the quantitative determination of prothrombin, several methods have been devised. The oldest is probably that of Howell.(1). This method, however, is no longer used by modern workers, probably because the prothrombin time as obtained by this method is influenced not only by the amount of prothrombin in the plasma but also by the other elements in blood coagulation, with the possible exception of thromboplastin. Several other methods have been recommended by various authors. Armand J. Quick(2), some 5 or 6 years ago, introduced a simple technique for use in the clinics About the same time, Smith(5) Warner, and Brinkhouse developed a two-stage titrimetric procedure which, according to the authors, "is an exact measure of prothrombin sufficiency or deficiency but is too complex for routine clinical work." Accordingly, they recommended a modified bedside technic. Micromethods based on the

foregoing tests have also been developed including the micro-method of Karabin and Anderson(6) based on Smith's method. Both micro-methods make use of blood obtained by deep skin punctures of the heel, finger, or ear and are suitable for prothrombin determinations in infants where it is very difficult to obtain blood from the vein.

The most widely used of the aforementioned technics are those of Quick and Smith and his associates. Both methods are simple but have their drawbacks. Quick(8) for example claims that the bedside technic of Smith, Warner, and Brinkhouse does not measure accurately the clotting time. On the other hand, Smith and his associates(8) claim that Quick's technic is more complex in the sense that plasma is recalcified and treated by thromboplastin, a centrifuge and water bath are needed; the amount of calcium is arbitrary and this has been found to affect the results of the test. Holmboe and Holmboe(9) further raise the following objections to Quick's test. The reagents must be made up fresh for each determination, the concentration of thromboplastin is variable and the normal time value for the method is too short, thus favoring errors in reading and preventing the measurement of prothrombin levels above normal. To overcome these objections, they performed a series of preliminary experiments using brain extract of different animals (rat, guinea pig, rabbit, sheep, and man) and varying concentrations of thromboplastin, plasma and calcium chloride. The experiments were based on Quick's method. After a series of such preliminary investigations, they have evolved a modified technique which they think has retained the good points of the original Quick's test and has at the same time overcome its defects. The technique is as follows:

Venous blood with 1:10 concentration of 0.1M sodium oxalate is centrifuged at high speed for fifteen to twenty minutes to separate the plasma from the cellular elements and tissue debris. 0.2cc. of plasma is mixed in a serology tube with 0.2 cc. of thromboplastin (diluted 1:5 with normal salt sol.) and incubated at 37°C. in a water bath for seven minutes after which the tube is removed from the water bath, the outside dried and 0.2cc. of 0.8M $CaCl_2$ immediately added. The tube is shaken gently, then slanted and rotated at fifteen second intervals. The interval between the addition of $CaCl_2$ and the time when the mixture first begins to show a fibrin web after gentle periodic rotation is taken as the prothrombin time. The thromboplastin solution is prepared from fresh sheep

brain. This is washed in water, the meninges and blood vessels carefully removed, cut into small pieces and dried with acetone. Drying is facilitated by repeated additions of acetone and grinding in a mortar. The macerated brain is freed of acetone and dried in the open air. The dry powder is extracted repeatedly with acetone until the precipitate forms with the addition of acetone. The other extract is precipitated by acetone and centrifuged to separate the precipitate. This is dried in air. It is a light brown waxy substance. 0.25 gm. of the substance is emulsified in 50cc. of normal salt solution and aqueous merthiolate (1:100,000) added as a preservative. The suspension is warmed to 56.5°C. for 10 minutes, with constant shaking. A homogenous opalescent mixture is thus obtained which keeps for as long as 3-5 months. The solution used in the test is a 1:5 dilution of the stock solution. The authors have also tried using rabbit's brain and obtained practically the same results as with sheep's brain. The normal values of prothrombin time obtained by the authors varied from 60-80 seconds with an average of 70 seconds. The results can also be expressed in prothrombin percentage, assuming 0 seconds to be 100%.

A few points have to be observed closely. The 1:10 concentration of sodium oxalate in the preparation of the plasma should be adhered to. Violent shaking of the mixture after the addition of $CaCl_2$ is to be avoided as this delays clotting. Badly hemolyzed blood is better discarded as gross hemolysis has been found to shorten the prothrombin time appreciably.

The first part of this study aims at establishing the normal prothrombin time among adult Filipinos by the technique of Holmboe and Holmboe using rabbit's and human brain. For this purpose, samples of blood were taken from 76 patients in the Medical and Surgical Services for the Philippine General Hospital who showed no clinical indications of any prothrombin deficiency. Some of these patients were suffering from certain blood diseases and therefore showed some hemorrnages, explainable by a defect in their blood platelets. A few of them had jaundice but did not manifest any hemorrhages even after surgical intervention.

Thromboplastin solutions prepared from both human and rabbit brain were used in all the case for purposes of comparison. The prothrombin time of the 55 samples, as determined with thromboplastin from rabbit's brain ranged from 45-80 seconds. In only one instance, however, was it found to be 80 seconds; this was a

case of pneumonia. Sixty-seven per cent of the cases gave a prothrombin time of 45 to 60 seconds. The mean normal value is 60.26 seconds. Prothrombin time determined by using thromboplastin from human brain was in a few instances longer by 5 seconds that that determined wiht thromboplastin from rabbit's brain. This tallies with the work of Holmboe and Holmboe who, using Quick's method, got an average prothrombin time of 32 seconds with rabbit's brain extract and 72 seconds with human brain extract.

The next part of the study was the determination of the concentration of prothrombin in the plasma which would appreciably lengthen the prothrombin time. The concentration of prothrombin in the plasma was varied by making several dilution of the plasma, viz., 1:2, 1:3, 1:4, 1:5, 1:6 and 1:7, and comparing the prothrombin time of the diluted plasma with the undiluted one. In 6 samples, this was done using both human and rabbit brain extract. In 7 samples, it was done using only human brain extract due to temporary shortage of the rabbit brain extract. The results of the experiment using rabbit brain do not parallel those of the experiment using human brain. In all 7 instances, clotting was delayed at a lower dilution when human brain was used than when rabbit's brain was used. Using thromboplastin prepared from rabbit's brain there was an increase in the prothrombin time of 14 2/7%-20% at a dilution of 1:4 (25% concentration of plasma prothrombin) in 3 cases, and of 13%-33⅓% at a dilution of 1:5 (20%) in 3 cases. This shows that the plasma prothrombin level has to fall to about 25% or less of the normal to produce any appreciable change in the prothrombin time. If, to start with, the plasma prothrombin level of an individual during health is high, a decrease of about 25% in the course of illness may not delay the clotting above the upper limit of normal prothrombin time (75 sec.). For example, individual A has a plasma prothrombin time of 45 sec. during health and B 76 seconds. Should both individuals suffer from any disease that will decrease the prothrombin level in their blood to 25%, and therefore increase the prothrombin time by say, 20%, B will have an abnormal protrombin time of 85 seconds while 3 will have a prothrombin time of 55 seconds which is still within normal limits. These results will give the impression that individual A is not suffering from any prothrombin deficiency when, in fact, he is. In view of this observation, we have decided to determine the prothrombin time at different plasma dilutions.

CLINICAL APPLICATION

The importance of the determination of the plasma prothrombin level cannot be over-emphasized. In adults, it is especially useful in cases of obstructive jaundice, biliary fistula, and diseases of the liver parenchyma where the patients show a tendency to bleed. Much investigation has been done by several workers from different parts of the world to explain the mechanism of the bleeding tendency in diseases of the biliary tract. Most workers believe that the hemorrhagic tendency is due to inability of the liver to maintain the plasma prothrombin at the normal level. This may be due to a defect in the liver cells themselves, which are supposed to be concerned in the manufacture of prothrombin, or to a lack of vitamin K, which is presumed to be necessary for the formation of prothrombin. Exactly how vitamin K acts is still a matter of conjecture. A vitamin K deficiency may in turn be due to an inadequate intake of the vitamin or to a defective absorption from the intestines. The absorption of vitamin K is influenced by the amount of bile in the intestines, it being a fat soluble vitamin. Thus, in obstructive jaundice and biliary fistulas, where little or no bile passes into the intestines, there is an impairment of vitamin K absorption, resulting in a vitamin K deficiency. Hence, the tendency to hemorrhages in such diseases. In portal cirrhosis and other liver diseases, the bleeding tendency is due to a defect in the liver cells themselves. Like any rational therapy, therapy with vitamin K is best guided by repeated determinations of prothrombin time. Especially is this so in cases that require surgical intervention. Prothrombin time should be done before operation to establish the necessity of a pre-operative course of vitamin K and to determine when the patient may be operated on with the least danger of hemorrhage incident to prothrombin deficiency. After operation, especially where artificial biliary fistulas have been produced, repeated prothrombin time determination is necessary to tell just how far and for how long vitamin K should be administered. It is our intention to extend our work further in this direction.

In the course of this study, we have come across 6 cases showing a prolongation of the prothrombin time. In all of them there was marked jaundice and tendency to hemorrhages. Two cases were of carcinoma of the head of the pancreas producing obstruction

of the common bile duct, 2 of cirrhosis of the liver, one of choledcholithiasis, multiple, and one of malignancy of the liver. In 4 of these cases, there was a favorable response to vitamin K therapy. The hemorrhages stopped and the prothrombin time returned to normal. In one case, vitamin K could not be tried because the patient insisted on being taken home. In the other case, vitamin K was administered but the patient died, not so much from the hemorrhages as from hepatic insufficiency.

In infants, determination of prothrombin time is important during the newborn period when the incidence of hemorrhagic diseases is high. Such hemorrhages are also believed to be due to vitamin K deficiency.

SUMMARY AND CONCLUSIONS

1. Determination of prothrombin time among 55 adult Filipinos showing no clinical evidence of prothrombin deficiency was done following the method of Holmboe and Holmboe using rabbit's brain and human brain.

2. The prothrombin time was found to be slightly longer when using human brain extract than when using rabbit brain extract on the same sample.

3. With rabbit's brain, the average normal prothrombin time in the 55 samples tested was found to be 59.54 seconds. The shortest time was 45 seconds and the longest, 60 seconds, with 67% giving values ranging from 45-60 seconds.

4. An appreciable change in the plasma prothrombin time occurred when the plasma concentration was reduced to 25%-20% when using rabbit's brain extract and 33⅓%-25% when using human brain extract.

5. The clinical value of prothrombin time determination is briefly presented.

BIBLIOGRAPHY

1. Todd, James Campbell and Sanford, Arthur Hawley, Clinical Diagnosis by Laboratory Methods, 7th edition, p. 230. 1932.
2. Quick, Armand J., The Nature of the Bleeding in Jaundice, J. A. M. A. v. 110, 1658, May 14, 1938.
3. Warner, E. D., Brinkhouse, K. M., and Smith, H. P., A Quantitative Study on Blood Clotting; Prothrombin Fluctuations under Experimental Conditions, Am. J. Physical, v. 114, p. 667, Feb. 1936.

4. (a) SMITH, H. P., ZIFFREN, S. E., OWEN, C. A. and HOFFMANN, G. R., Clinical and Experimental Studies on Vitamin K, J. A. M. A. v. 113, p. 380, July 29, 193.

(b) ZIFFREN, S. E., OWEN, C. A., HOFFMANN, G. R. and SMITH, H. P., Control of Vitamin K Therapy; Compensatory Mechanisms at Low Prothrombin Levels, Proc. Sec. Exper. Biol. & Med. v. 40, p. 595, April 1939.

5. KATE, KATSUI and PONCHER, HENRY, G. Prothrombin in the Blood of Newborn Infants, J. A. M. A. v. 114, p. 749, March 2, 1940.

6. KARABIN, JOHN, E. and ANDERSON, EDWARD R.: A Simplified Micro Test of Plasma Prothrombin, Jour. Lab. and Clin. Med., v. 26, p. 723, January 1941.

7. DAM, HFARIK, and CLAVIND, JHANNESS Vitamin K in Human Pathology. Lancet, v. 1, p. 720, March 26, 1938.

8. OLSON, K. B. and MENSEL, H.: Bleeding Tendency in Obstructive Jaundice; Correction by Means of Vitamin K, Surgery, v. 6, p. 206, Aug. 1939.

9. See 4(b).

10. HOLMBOE, H. S. and HOLMBOE, R. W.: Modified Technique for Prothrombin Determination in Blood, Jour. Lab. and Clin. Med., v. 26, p. 408, November 1940.

11. SCANLON, G. H., BRINKHOUSE, K. M., WARNER, E. D., SMITH, H. P., and FLYNN, J. E.: Plasma Prothrombin and the Bleeding Tendency, J. A. M. A., v. 112, p. 1898, May 13, 1939.

TABLE I.—*Prothrombin Time Using Rabbit's Brain*

Dilution of Plasma	Prothrombin Time in Seconds						Remarks
	1	*2*	*3*	*4*	*5*	*6*	
100%	60	70	75	45	50	60	
1:1 (50%)	60	70	75	50	50	60	
1:2 (33 1/3%)	60	70	75	50	55	65	
1:3 (25%)	70 (16 2/3%)	80 (14 2/7%)	75	50	60 (20%)	65	14 2/7-20% (4 cases)
1:4 (20%)	90	—	85 (13 1/3%)	60 (33 1/3%)	75	75 (20%)	13 1/3-33 1/3% (3 cases)
1:5 (16 2/3%)	95	—	85	75	75	80	
1:6 (14 2/7%)	110	—	—	80	90	120	

TABLE II.—*Prothrombin Time Using Human Brain.*

Dilution of Plasma	Prothrombin Time in Seconds													Remarks
	1	2	3	4	5	6	7	8	9	10	11	12	13	
100%	75	60	75	45	50	55	65	65	65	60	55	60	50	
1:1														
50%	75	65	75	55	50	60	70	70	65	65	60	70	55	
1:2	75	65	80	60	55	65	80	75	70	80	65	75	60	15-33 1/3%
33 1/3%				33%		18%	23%	15%		33%	18%	25%	20%	(8 cases)
1:3	90	75	100	75	60	75	120	80	75	85				15-33 1/3%
25%	20%	25%	33%		20%				15%		70	85	60	(5 cases)
1:4														
20%	95	95	105	90	70	75	240	85	85	100	75	95	75	
1:5	95													
16 2/3%		125		100	90	120	360	90	110	105	75	100	80	
1:6														
14 2/7%	120	210		120	100	120	390	110	130	105	85	120	90	

LEUKEMIC RETICULOENDOTHELIOSIS IN THE PHILIPPINES

BY

EUGENE STRANSKY, M.D., ERNST J. HIRSCH, M.D., and
HILARIO ZIALCITA, M.D.

College of Medicine, Department of Pediatrics,
University of the Philippines

Leukemic reticuloendotheliosis in the Philippines was first discussed by Stransky and Regala in one of the Staff Conferences of the Philippine General Hospital in June 1941. The authors gave a full report of the case to the Convention of the Philippine Medical Association in December 1943. Since then four additional cases were diagnosed and a fifth observed. This gives us the opportunity of offering a more detailed description of this rare variety of leukosis.

The first case of this kind of leukosis was published by Reschad and Schilling(1) in 1913 under the title "A New Kind of Leukemia Due to Genuine Transitional Cells (Splenocytic Leukemia) and Its Importance for the Individuality of These Cells." Fleischmann(2), Bingel(3), Ewald(4), Letterer(5), Krahn(8) described similar cases although their nomenclature varied.

In the United States, probably the first described case was that of Rosenthal(7), although he considers monocytic leukemia merely a variety of myelosis and denies it as an entity. Farley(8) and Dameshek(9) labelled their cases monocytic leukemia. In 1942 Evans(12) described the latest developments and cited an extensive bibliography.

What is "leukemic" and what is "aleukemic" reticuloendotheliosis? Leukemic reticuloendotheliosis means monocytic or histiocytic leukosis (leukemia), while aleukemic reticuloendotheliosis means a generalized proliferation of the reticuloendothelial cells without a leukemic blood picture. In aleukemic reticuloendotheliosis, there is severe anemia, marked hepatosplenomegaly, lymphadenopathy, hemorrhagic diathesis and high fever, all symptoms pointing to acute leukosis. The blood picture is, however, normal. As mentioned

before, Letterer(5) coined the words "aleukemic reticulosis." Podvinec and Terplan(13) call the disease "reticular hyperplasia" due to an infection of unknown origin; Siwe(14) calls it "acute aleukemic reticuloendotheliosis." Since the publication of Abt and Denenholz(15) the syndrome has been called Abt-Letterer-Siwe's disease. Glanzmann and Walter(16) stressed the fact that "infectious" reticuloendotheliosis is closely connected with storage diseases. They reported a child who developed hepatosplenomegaly, generalized adenopathy, petechial bleedings in the skin, and high fever, without any characteristic blood changes. Later the typical symptoms of cholesterol storage disease (Hand-Schueller disease) were observed. A careful histological study of the different organs showed the close connection between infectious (aleukemic) reticuloendothelosis and storage diseases.

Sternberg(17), (18), forty years ago thought that lymphosarcoma and acute lymphadenosis (lymphatic leukemia) were closely related. Isaacs(19) found 15 cases among 43 cases of lymphosarcomatosis, with a terminal blood picture and clinical symptoms similar to acute lymphadenosis. He called his cases "lymphosarcoma cell leukemia." Apitz(20),(21) published several papers on leukosis as malignant new growth. A critical study on the close relation between myelosis and malignant newgrowth was made by Moeschlin and Rohr(22) with an extensive bibliography.

On the other hand there may be leukemia changes or metaplasias of certain organs without generalization of the leukemic process. These cases may be compared, according to Roessle to localized malignant newgrowth. Hicling(23) wrote in 1937: "Many cases have been described of patients with massive enlargements of the spleen due to myeloid metaplasia, in whom examinations of the circulating blood have not revealed the characteristic blood picture of myeloid leukemia." He calls these cases "chronic, non-leukemic myelosis." There are not less than 27 cases collected which have been autopsied. In 11 cases, myeloid metaplasia was found in the liver, in 6 cases, hyperplasia, as in chronic myelosis, in the bone marrow, while in 3 cases there was myeloid metaplasia of the lymph nodes. There is no doubt, therefore, that myeloid metaplasia may take place in the different organs even without symptoms of chronic myelosis in the blood.

Stransky and Quintos(24) observed two cases of myeloid and one of lymphatic metaplasia of the spleen by splenic puncture. The

leukemic infiltration was restricted to the spleen, no other organs, nor blood nor bone marrow were affected. We must of course not forget chloro-leukemia, which is definitely a link between acute leukosis and malignant tumors, and furthermore that cases are known which developed tumor-like periostal infiltrations without the green color of chloroleukemias.

Leukemic reticuloendotheliosis, like other leukoses, can be acute or chronic. Almost all cases reported in the literature belong to the acute type. The chronic cases are extraordinarily rare. Acute leukemic reticuloendotheliosis is hematologically sometimes different from the acute myelosis and lymphadenosis. In acute myelosis and lymphadenosis we observe, as a rule, the so-called hiatus leukemicus, a preponderance of the blast cells (60-99% of all white cells are blast cells), and absence of almost all intermediary stages of cell forms with the exception of few mature cells. In leukemic reticuloendotheliosis the percentage of the immature reticuloendothelial cells may be low and the hiatus may be absent.

In 1939 Schultz and Krueger(25) gave an excellent survey of monocytic leukemia, describing the different cells in this disease. They distinguish three well-defined types of cells of reticuloendothelial origin in the peripheral blood: (1) Large cells with a diameter of at least twenty, often thirty and sometimes even of fifty microns, with a large nucleus of reticuloendothelial structure, containing several nucleoli, having basophilic, at times dark blue cytoplasm. The nucleus is spheroid, rather big in comparison with the size of the cytoplasm. As myeloblasts and lymphoblasts are never so large and the nuclear structure is quite different, it is not at all difficult to differentiate this type of reticuloendothelial cells from other blast cells. (2) There are larger and smaller reticuloendothelial cells in the blood, in all stages of maturation, monoblasts, promonocytes and monocytes, containing lobulated, nearly segmented nuclei, but in spite of the lobulation, the nuclear structure is immature. It seems that these cases are either an aberrant or a precipitated maturation of the nuclei. These cells can be compared with the paramyeloblasts of Naegeli(26),(27) and Hittmair(28) considers these cells as myeloid cells and deny their monocytic nature. (3) The cells have exactly the same structure as in(1) the difference being only in the size; the cells are smaller than the myeloblasts, that is 10-16 micra in diameter. There are numerous more dif-

ferentiated cells, the so-called promonocytes and even mature monocytes, which facilitate the differentiation of these cells. Bykowa(10) observed a case with only small cells, which were difficult to differentiate from lymphatic cells. Supravital staining, however, was of great help in these dubious cases, as reticuloendothelial cells stained with neutral red look quite different in shape and structure from myeloid and lymphoid cells. Unfortunately war conditions did not allow us to use supravital staining in our cases. The peroxydase test is, compared to the supravital staining with neutral red, of no use in the differentiation of reticuloendothelial and myeolgenous cells, as cells of both, having reached a certain stage of maturity, are peroxydase positive; the so-called stem cells are invariably peroxydase negative, similar to lymphoid cells in all stages of development.

According to Hittmair(28) "monocytic leukemia is the most interesting, but simultaneously the most difficult and most complicated chapter of hematology." He differentiates two types of this kind of leukosis, the so-called monocytic leukemia, which is due to the hyperplasia of the myeloid system and leukemic reticuloendotheliosis, characterized by a hyperplasia of the reticuloendothelial system, not observed in the first type. Hittmair shares the opinion of Naegeli(59) to a certain degree. He assumes that there are cases of leukosis with monocytoid cells of bone marrow origin. As these cells are not real reticuloendothelial cells, there is no hyperplasia of the retitculoendothelial apparatus and the anatomical changes are those of a myelosis. He calls these cases the Naegeli-type, while the second, with marked changes in the reticuloendothelial apparatus, are the Schilling-type of monocytic leukemia. According to him there are mixed forms, where both types are present, a hyperplasia of the myeloid and reticuloendothelial system together. As the mesenchyme is considered by Downey(11) Hittmair(28) and many others as a pluripotent cellular system, which may develop in different directions, the mesenchymal cell should be considered the mother cell of all kinds of blood cells. These primordial cells may develop to red as well as to different kinds of white cells. This theory may explain why erythroblastosis is so frequent in leukemic reticuloendotheliosis. It does not explain, however, the fact that in leukemic reticuloendotheliosis, as already mentioned, the leukemic infiltrations are frequently limited. Here, the theory of the tumor-like character of the disease gives a pos-

sible clue. Developing this idea, we reached a new point of view for the unitary etiology of leukoses. A strong stimulus on the mesenchyme, the nature of which is so far unknown, is the common cause of all leukoses. It is not clear why the same stimulus leads to a myelogenous, lymphatic or reticuloendothelial reaction. But, apparently, one kind of leukosis can change to another. In acute myelosis and acute lymphadenosis, erythroblastosis is absent. Therefore leukemic reticuloendotheliosis may be considered the most primitive and embryonic form of all leukoses.

In leukemic reticuloendotheliosis there is as a rule no generalized involvement of the reticuloendothelial apparatus. In myelosis and lymphadenosis, generalized involvement of the respective tissues is a constant phenomenon. In leukemic reticuloendotheliosis, the causative stimulus may be selective; in other leukoses it is generalized. This would mean a link between leukosis as a whole and malignant newgrowth.

We have considered it necessary to discuss the etiology of leukemic reticuloendotheliosis in connection with other diseases of the reticuloendothelial system and the leukoses so that the clinical symptomatology and the hematological particularities of this disease can be better understood.

CASE REPORTS

Our first case, a thirteen-year old girl, was previously described in a paper by Stransky and Regala(68) submitted for publication in 1941 to the Acta Medica Philippina. The important data are summarized below.

T. Q. was admitted May 28, 1941 on account of general body weakness, pallor and vaginal bleedings. The girl had her menarche the last day of January, 1941. In February, the menses were normal; in March, there were no menses at all. In April vaginal bleeding started at the expected time, did not stop, and developed to a profuse metrorrhagia. The blood examination revealed a white cell count of 65,000 per cc. The diagnosis of myelogenous leukemia was made and blood transfusion given, which stopped the vaginal bleedings. Past history irrelevant; no blood diseases in the family.

On admission the child was extremely pale, slightly febrile, with few pinpoint hemorrhages all over the skin and conjunctiva. Physical findings were normal, no enlargement of spleen and liver. Lymph nodes were all of normal size.

The girl stayed eight weeks until her death. She ran a continuous temperature of 38-39 degrees centigrade. One week before death she developed multiple skin and retinal bleedings. At this time she complained of abdominal pain; four days previous to death she had loose bowel movements. The blood examinations were as follows:

Date	29/5/1941	6/6/1941	30/6/1941	6/7/1941
Hemoglobin	23% = 3.6 gm.	—	24% = 3.75 gm.	—
Red cell count	960,000	1,040,000	1,490,000	1,050,000
White cell count	16,800	7,950	3,650	5,700
Nucleated red cells	42:250	90:250	47:250	49:250

Differential count:

	B	E	Mbl	Prom	M	J	St	S	L	RE Mobl	RE Prom	RE Mo	RE End
29/5/41	—	—	—	0.4	2.4	6.0	10.0	29.2	11.6	30.0	2.4	7.6	0.4
31/5/41	—	—	—	—	1.2	3.2	10.4	26.0	18.0	31.6	4.8	3.6	1.2
2/6/41	—	—	—	—	0.8	4.0	10.8	30.8	14.8	30.0	5.2	2.8	0.8
6/6/41	—	3.2	—	—	—	2.4	4.0	28.8	24.0	22.0	4.0	3.2	0.4
9/6/41	—	—	—	—	0.4	0.8	4.4	26.0	40.0	24.4	2.4	1.2	0.4
17/6/41	—	2.0	—	—	—	0.8	2.8	13.2	35.2	34.0	8.0	4.0	—
30/6/41	—	2.4	—	—	—	0.4	3.2	8.8	46.8	32.0	4.0	2.4	0.4
6/7/41	—	—	—	—	—	1.6	3.6	4.8	44.0	37.6	5.6	2.8	0.4
Bone marrow:													
29/5/41	—	0.8	2.0	4.0	12.0	23.2	11.6	6.8	4.0	30.4	4.8	0.8	—
17/6/41	—	0.8	—	0.8	3.6	9.2	7.6	5.2	1.6	40.0	4.2	2.0	—

Bone marrow:	Cell count	Normoblasts	Basophilic erythroblasts	Makroblasts	Proery-throblasts	Mitotic red cells	All red cells
29/5/41	88,000	153	24	23	—	—	200:250
17/6/41	69,800	325	80	28	5	—	438:250

There is no niatus leukemicus either in the peripheral blood, nor in the bone marrow. The monocytic cells (monoblasts, promonocytes, monocytes and typical big reticuloendothelial cells) never exceeded 40-80% of the total white cell count. The monocytic cells were very large, about 30 micra in diameter with a bluish cytoplasm, a relatively big, vesicular nucleus containing several nucleoli and with fine reticular structure of the nuclear chromatin. Erythroblastosis, both in peripheral blood and bone marrow was a constant and striking feature. The diagnosis of leukemic reticuloendotheliosis was confirmed by the post mortem examination. Leukemic infiltrations were found in the liver, lungs, kidneys, and spleen, while the lymph glands were only slightly infiltrated or not at all. The type of the infiltrating cells was exactly the same in the blood and bone marrow. At autopsy, amebic ulcers were found in the intestines. One of them had perforated and a peritonitis developed. The case was presented to a Staff Conference of the Philippine General Hospital. As our diagnosis was not unanimously accepted, blood and bone marrow smears and histological sections of the different organs were sent to Dr. Downey of Minneapolis, who was kind enough to check our findings. He confirmed our diagnosis and wrote that the amebic dysentery had nothing to do with the blood dyscrasia. The disease had lasted three months.

The second case was a two-year old male child. He was brought to the hospital almost agonizing, very pale, with petechial hemorrhages all over the skin, hepatosplenomegaly and generalized enlargement of the lymph nodes. The disease started a few weeks previous to admission with fever and purpuric spots. The child stayed only a few hours in the Hospital, was taken home against advice and died few hours later. We were, however, able to examine the blood carefully.

Red cells ..... 1.820,000 per cmm. blood
White cells ..... 440,000 per cmm. blood

										R.E.		
	B	E	Mbl	Prom	M	J	St	S	L	Mobl	Prom	Mo
Differential count:	—	0.8	0.8	0.8	2.0	4.0	6.0	14.8	2.8		68.6	

Normoblasts	43
Basophilic erythroblasts	23
Macroblasts	19
Proerythroblasts	13
Mitotic red cells	2

100:250 or 40:100 nucleated red cells to white cells in the peripheral blood.

The cells were similar to those observed in the first case, very large monocular cells, much bigger than myeloblasts or lymphoblasts. There were few monocytoid cells with aberrant maturation of the nucleus, which we considered as promonocytes and monocytes. Even these were markedly larger than normal monocytes. There was also a striking erythroblastosis, even more marked than in the first case, with the most immature elements of the erythron series and some immature cells of the myelon series. There were not less than 125,000 nucleated red cells in one cc. of blood, a point which is never observed in acute myelosis or lymphadenosis. Unfortunately, a bone marrow examination could not be done. There were hepatosplenomegaly and generalized enlargement of the lymph nodes, similar to acute myelosis and lymphadenosis.

Case 3, hospitalized in St. Luke's Hospital, Manila, came to the attention of the senior author upon request for consultation. This was a 14-year old girl. The illness started in December 1943 with fever, general weakness and extreme pallor. On account of the enlarged heart, distinct systolic murmur and fever, the diagnosis of the attendant physicians was endocarditis lenta. The first blood examination was done by us in the last day of December 1943. There were 640,000 red cells per cc. blood, while the white cells amounted to only 2,200. Not more than 9% immature reticuloendothelial cells were seen in the blood smear. There was no hepatosplenomegaly, no enlargement of the lymph nodes, no nucleated red cells in the peripheral blood. In February 1944 a bone marrow examination was done in our laboratory. There were more than 98 per cent of immature reticuloendothelial cells and only few granulocytes and lymphocytes. The cells, contrary to the former two cases, were with very few exceptions, not bigger than the monocytes (12-18 micra), but with exactly the same nuclear structure as described in the other two cases. There were only a few more mature monocytoid cells of the same size. The patient developed hemorrhages from the gums from time to time and the anemia remained unchanged in spite of repeated blood transfusions. One of us saw the patient July 1st 1944, three days previous to death. Blood examination revealed:

B	E	M	J	St	S	L	Mobl	Prom	Mo	
										1:250 nucleated red cells.
—	—	—	2.0	11.6	23.2	19.6	40.8	2.0	0.8	
								43.6		

The percentage of the reticuloendothelial cells had risen to more than 40%. The character, size and shape of the cells as compared with the findings in December 1943 were unchanged. The patient died with symptoms of severe hemorrhage on July 4, 1944.

Case 4 is a 23-year old female, admitted to the Philippine General Hospital April 23rd, 1944. She died May 9th, 1944. Three weeks previous to admission, she developed cough with slight fever. A few days later, headache, chills, general malaise, with higher temperature, were noticed. Several days previous to admission, dyspnea, chest and back pains aggravated the picture. On admission the liver was found to be four fingers below the right costal arch, the spleen reached the umbilical line. During her stay in the Hospital, a continuous remit-

tent fever was noticed, at first up to 39°C., later around 38°C. rising before death to 40°C. One week before death a pustular eruption appeared on the skin of the back, chest and face. Widal test, blood cultures and examinations for malaria were made and found negative. The feces showed many ascaris eggs. As the patient was distinctly jaundiced, bilirubin determination was done in the laboratory of the Department of Medicine. The results showed indirect bilirubin 0.417, direct bilirubin 3.948 mg. Takata-test ++++.

Peripheral blood:	*24/4/1944*	*29/4/1944*	*2/5/1944*	*8/5/1944*
Red cell count	4,200,000	—	3,620,000	3,500,000
Hemoglobin	—	—	12.2 gm.	—
White cell count	10,830	11,500	7,500	7,350
Platelet count	108,000	Reticulocytes: 2.6%		

Differential count:	B	E	M	J	St	S	L	R.E.	
	—	—	—	—	4.0	42.8	12.0	40.4	No nucleated red cells.

Bone marrow, total cell count extremely low (around 20,000).

Differential count:	B	E	Mbl	Prom	M	J	St	S	L	R.E.
	—	1.2	1.6	3.2	4.0	6.4	6.8	7.2	5.2	64.8

Normoblasts	22	
Basophilic erythroblasts	9	37:250 or 14.8:1000 nucleated red cells
Makroblasts	1	to white cells,
Proerythroblasts	none	
Mitotic red cells	none	

Megakaryocytes very few.

All reticuloendothelial cells in the blood and bone marrow were very large cells with a diameter of 30-35 microns containing a big spheroid nucleus with few vacuoles and a fine reticular structure. There were very few more mature monocytoid cells. No signs of anemia in the peripheral blood; the erythropoietic activity of the bone marrow was very poor. Although the cell count in the bone marrow was strikingly low, a reticuloendothelial metaplasia was apparent as the percentage of the reticuloendothelial cells was very high.

Very revealing were the findings in the aspirated splenic juice. The cell count was extremely high. The smear looked like the blood smear of a leukosis with around 100,000 cells per cc. blood. The differential count:

								R.E.			
	B	E	M	J	St	S	L	Mobl	Prom	Mo	Mit
	—	—	—	1.6	4.6	10.4	8,4	71.6	1.2	0.8	0.4
									74.0		

Normoblasts	10	
Basophilic erythroblasts	5	16:250 or 6.4:100 nucleated red cells
Macroblasts	1	to white cells.
Proerythroblasts	none	
Mitotic red cells	none	

From these findings we deduced a marked hematopoietic activity of the spleen. Simultaneously, there was an erythropoietic activity, as nucleated red cells were found in the aspirated splenic juice, while, in the peripheral blood, there were no nucleated red cells. As there were many more cells in the splenic juice than in either the peripheral blood or bone marrow, we considered this the direct evidence, intra vitam, of leukemic metaplasia of the spleen. The overwhelming majority of the cells in the spleen were large, immature reticuloendothelial cells, similar to those in the peripheral blood and the bone marrow.

The most important autopsy findings were as follows: Spleen very large, double the normal weight; the color dark red, soft, the pulp on section greyish, easily scraped off. Malpighian corpuscles not prominent. Liver, weight more than double, (over 2,500 gm.) surface smooth, no cirrhosis, very yellow in color, not like jaundice, but distinctly the yellow color of fatty degeneration. Consistency soft. Bile ducts normal. Lymph nodes not enlarged, except the lymph follicles on the base of the tongue. Tonsils normal. Moderate bleeding along the site of the sternal puncture, no other symptoms of hemorrhage. Sternal marrow red, that of the femur pinkish in color.

Histological sections: Spleen under the low power markedly cellular, large numbers of red cells, Malpighian corpuscles not prominent. Under high power, the cells looked large with big vesicular nuclei and definite cytoplasm. Some of the detached and enlarged cells showing phagocytic activity were hyperlastic, reticuloendothelial cells of the spleen. No signs of malarial pigmentation. Liver: extensive fatty degeneration, which partially accounts for the enlargement and yellow color. There were collections of cells in the different parts of the liver, without relation to the porta hepatis; they were in the periportal area around the central veins. The cells were large, with a vesicular nucleus. They were found even in the sinuses. It seems that the Kupffer cells were also enlarged. A few necrotic patches of liver cells were found. No cirrhosis. No proliferation of the bile ducts. Skin: no leukemic or other cellular infiltrations. Except for the bone marrow (the same cell type as in the spleen and liver) no cellular infiltration in any other organ.

The findings in the spleen and liver, as well as in the bone marrow and peripheral blood are definitely in favor of leukemic

reticuloendotheliosis. The anemia was very slight, on account of the fact that the leukosis was not yet in a sufficiently advanced stage to produce anemia at the time the patient died.

The most instructive case in our observations is Case 5, a private patient, female, 22 years old. She came from Negros (one of the Visayan Islands) to Manila on March 1944 on account of body weakness and increasing pallor. Blood examination revealed the following:

Date	17/4/1944	28/4/1944	15/5/1944	27/5/1944	24/6/1944
Red cell count	3,420,000	2,500,000	2,460,000	—	1,330,000
White cell count	—	55,400	—	—	35,100
Nucleated red cells	18:250	none	32:250	8:250	6:250

								R.E.			
Date	B	E	M	J	St	S	L	Mobl	Prom	Mo	Pl
17/4/44	0.4	1.2	—	0.8	6.0	40.0	47.2	—	—	4.0	0.4
28/4/44	—	8.0	—	—	6.4	61.6	20.8	—	—	2.8	—
15/5/44	—	0.4	—	—	8.0	50.0	39.2	—	—	9.6	—
27/5/44	—	—	0.8	4.0	9.2	39.2	39.2	—	—	7.6	—
24/6/44	—	—	1.2	2.8	3.6	16.0	14.0	39.2	12.0	9.6	—
									60.8		

Bone marrow: May 15, 1944.—Cell count 66,400 per cc. aspirated juice.

Differential count:	B	E	Mbl	Prom	M	J	St	S	L	Mo
	—	0.4	7.2	9.2	11.2	18.8	10.4	24.0	17.6	1.2

Normoblasts	323	
Basophilic erythroblasts	105	543:250 or 227.2:100 nucleated red cells
Makroblasts	70	
Proerythroblasts	35	to white cells.
Mitotic red cells	10	

Megakaryocytes normal.

May 27—Reticulocytes (twice repeated): 2%.

At first glance the above findings, with the exception of the last, did not seem at all characteristic of leukemic reticuloendotheliosis. Likewise, the clinical symptomatology was not suggestive of leukosis. The patient was pale, weak, afebrile, without hepatosplenomegaly or lymphadenopathy. The diagnosis was obscure until we learned that just before we saw the patient a blood transfusion had been administered. We therefore considered the possibility that

the blood findings were obscured by the blood transfusion. A second blood examination was done. Now nucleated red cells were absent, although the former anisocytosis and polychromasia were still present. The third examination revealed an increasing anemia with a striking erythroblastosis; the monocytes had risen to 9.6%, but without immature cells. The fourth showed again a slight monocytosis with a few immature granulocytes and a constant erythroblastosis. We considered the possibility of hemolytic anemia. The erythropoietic hyperactivity of the bone marrow suggested a hemolytic process. However, the absence of any spherocytosis, the low reticulocyte count, absence of urobilinogenuria and normal bilirubin level in the blood serum were definitely against hemolytic anemia.

The patient was afebrile, though weak and bedridden, until the middle of June, when she developed high fever. The possibility of malaria, endocarditis lenta or septicopyemia was entertained by the attending physicians. But blood cultures were repeatedly negative and no malaria parasites could be found in the blood. A few days later the patient developed ulcerative stomatitis with marked enlargement of the cervical lymph nodes, pallor and dizziness. A blood transfusion brought no relief whatsoever. Another blood examination permitted a final diagnosis. The cells now found were large reticuloendothelial cells, almost all blast cells, but there was lobulation and even segmentation in several cells, without any perceptible change in the immaturity of the nuclear structure. There was no difficulty recognizing the cells as having a typical monocytic, reticuloendothelial origin. The patient became weaker and paler from day to day; the stomatitis, resisting all therapeutic measures, grew more severe and, one week after the last blood examination, she died.

This last case is of considerable importance in that it gave us the opportunity of observing an acute leukemic reticuloendotheliosis in the preleukotic stage. There are great differences of opinion as to how an acute leukosis starts. In our particular case, a marked anemia with erythroblastosis developed, probably months previous to the frank acute leukosis. This anemia may be due to an excitement of the reticuloendothelial system or to a disorder affecting the system, without any discharge of immature cells into the peripheral blood. The activated reticuloendothelial system was phagocytizing more red cells than normally; an anemia of he-

molytic type developed, but did not produce any jaundice, as long as the liver function was normal. In Case 4, due to a liver affection, jaundice developed, in contrast to Case 5. It is probable that the bone marrow is able to counterbalance the increased destruction of the red cells for some time. When nucleated red cells disappear from the peripheral blood, we have to assume that the maturation of the red cells goes on in the bone marrow without discharge into the peripheral blood. After the first anemic stage, without any change in the blood picture (white cells), there is a short stage of discharge of increased number of mature cells (in this particular case monocytes). Finally the clinically manifest disease sets in, with fever, ulcerative stomatitis, hepatosplenomegaly and leukotic blood picture. In Case 5 this manifest stage was of short duration, only 2-3 weeks. We think therefore that acute leukoses may have a long pre-leukotic stage (in our case characterized by anemia), which is difficult or impossible to diagnose, followed by a short intermediary stage of increase of the cells of the involved system, until the clinically manifest disease sets in. The process is apparently acute, but as a matter of fact, is long standing, with an acute terminal stage.

Endocarditis lenta due to streptococcus viridans may cause severe anemia, involvement of the whole reticuloendothelial apparatus, hemorrhages and hepatosplenomegaly. Two of our cases were at first diagnosed as such. In endocarditis lenta there is, as a rule, a leucocytosis (although some cases may have leukopenia), neutrophilia and marked shift to the left. There may be a few immature reticuloendothelial cells in the peripheral blood, and in the bone marrow there may be an increase of reticuloendothelial cells. These cells are, however, of the mature type and the predominating cells are, contrary to leukemic reticuloendotheliosis, granulocytes.

There are cases of so-called aleukemic or infectious reticuloendotheliosis, which may suggest, as already discussed, leukemic reticuloendothelosis. The blood picture, excepting a secondary anemia and infectious changes, is almost normal and therefore the differential diagnosis should not be difficult.

Stransky and Pecache(31) observed a case of Brill(32)-Symmer's(33) disease (giant follicular lymphoma), which developed later to lymphogranuloma malignum (Hodgkin's diseases). In this par-

ticular case there were immature reticuloendothelial cells in the peripheral blood, in the bone marrow and in the aspirated splenic juice. Their percentage was however low, and they disappeared from the blood and bone marrow during the development of the disease. There was a generalized enlargement of the lymph nodes and a marked splenomegaly. There was fever, but no hmorrhage. Leukemic reticuloendotheliosis was considered as a possibility, but discarded as soon as a histological section of an excised gland was done. Brill-Symmer's disease is a syndrome, common in the early stage of Hodgkin's disease, lymphosarcoma and acute lymphadenosis, and therefore is evidence of close relationship between the diseases of the lymphatic system.

SUMMARY

Five cases of leukemic reticuloendotheliosis are discussed, with presentation of the clinical and hematological features of the disease. The morphology of the cells characteristic of the disease is described. In the fifth case the pre-leukemic stage of the disease was observed.

REFERENCES

1. Reschad H. and Schilling-Torgau V. Ueber eine neue Leukaemie durch echte Uebergangsfermen (Splenozytenleukamie) und ihre Bedeutung for die Selbstaendigkeit dieser Zellen. Muenchn Med. Wochchr, LX:1981, 1913.
2. Fleischmann P. Der zweite Fall von Monozytenleukamie. Fol. Haemat. XX:17, 1915.
3. Bingel H. Monozytenleukamie? Deutsche Med. Wochschr. XLII: 1503, 1918.
4. Ewald O. Die leukaemische Retikuloendotheliose. Deutsch, Arch. Klin. Med. CXLVIIff222, 1923.
5. Letterer E. Aleukaemische Retikulose. Frankf. Zeitschr. Path. XXX:677 1924.
6. Krahn H. Retikuloendotheliale Antwort... Deutsch. Arch. Klin. Med. CLII:179, 1926.
7. Rosenthal N. Some atypical cases of leukemia. Med. Clin. North America. IV:1607, 1921.
8. Farley D. Monocytic leukemia. Med. Clin. North America, XIII:991, 1930.
9. Dameshek W. Acute, monocytic (histiocytic) leukemia; review of literature and case reports. Arch. Int. Med. XLV:718, 1930.
10. Bykowa O. Retikuloendotheliale Leukosen (mit Affektion der Haut). Fol. Haemat. LI:96, 1933.
11. Downey H. Monocytic leukemia. in Handbook of Heamotology. II:1273, 1938, (P. H. Hoeber).
12. Evans T. S. Monocytic leukemia. Medicine, XXI: 421, 1942.

13. PODVINEC E. and TERPLAN K. Zur Frage der sogenannten akuten, aleukaemischen Retikulose. Arch. Kindernelik. XCIII: 40. 1931.

14. SIWE S. Die Retikuloendotheliose, ein neues Krankheitsbild unter den Splenomegalien. Zeit chr. f. Kinderheilk. LV: 212, 1933.

15. ABT A. F. and DENENHOLZ E. J. Letterer-Siwe's disease; splenomegaly, associated with widespread hyperplasia of nonlipoid storing macrophages; discussion of the so-called reticuloendothelioses. Am. Journ. Dis. Child. LI: 498, 1936.

16. GLANZMANN E. and WALTER K. Infektiœse Retikuloendotheliose und ihre Bezienhungen zur Schueller-Christian'schen Krankheit. (Monographe), 1941 (Huber).

17. STERNBERG C. Pathologie der Primaererkrankungen der Lymphdrusen, und des haematopoetischen Apparates. Erg. d. allg. Path. VIII: 380, 1905.

18. STERNBERG C. Ueber Leukosarkomatose. Wien. Klin. Wchschr. XXI: 475, 1908.

19. ISAACS R. Lymphosarcoma cell leukemia. Ann. Int. Med. XL: 657, 1937.

20. APITZ, R. Die neoplastische Natur der menschlichen Leukosen. Virch. Arch. path. Anat. ICCC: 1, 1937.

21. APITZ, R. Dies neoplastiche Natur der menschlichen Leukosen. Med. Welt. XIV: 85, 1940.

22. MOESCHLIN I. and ROHR K. Klinische und morphologische Gesichtspunkte zur Auffassung der Myelose als Neoplasma. Erg. inn. Med. u. Kinderheilk. LVII: 723, 1939.

23. HICKLINGS R. A. Chronic, nonleukemic myelosis. Quart. Journ. Med. VI: 253, 1937.

24. STRANSKY E. and QUINTOS F. N. On the diagnostic importance of splenic puncture. Proc. ann. conv. Phil. Med. Ass. 1943.

25. SCHULTZ W. and KRUEGER E. Monozytenleukaemie. Erg. Inn. Med. u. Kinderheilk, LVI: 56, 1936.

26. NAEGELI O. Blutkrankheiten und Blutdiagnostik. V. Edition, 1931 (Springer J.)

27. NAEGELI O. Rotes Knochenmark und Myeloblasten. Deutsche Med. Wochschr. XXVI: 287, 1900.

28. HITTMAIR A. Die Monozytenleukamie und die leukamische Retikuloendotheliese. Fol. Haemat. LXVI: 1, 1942.

29. STRANSKY E. and REGALA A. Leukemic reticuloendotheliosis. Rep. conv. Phil. Med. Ass. 1943.

30. STRANSKY E. and PECACHE L. V. On malignant newgrowths in childhood. Ueber kongenitale Leukemie. Montatsschr. f. Kinderheilk. XXIX: 654, 1925. Ann. Paediatr. (in print).

31. BRILL N. E., BAEHR G. and ROSENTHAL N. Splenomegalic, lymphatic hyperplasia generalized, lymph follicle hyperplasia of the lymph nodes and the spleen. Trans. Ass. Am. Physicians. XXXIX: 371, 1924.

32. SYMMERS, D. Giant follicular lymphadenopathy, with or without splenomegaly, . . . Arch. Path. XXVI: 603, 1948.

THE RELATIVE VALUE OF THE ACID-ETHER CENTRIFUGATION AND FAUST-MELENEY EGG-HATCHING TECHNICS IN THE DIAGNOSIS OF SCHISTOSOMIASIS JAPONICA *

T. P. PESIGAN, M.D., M.P.H., and M. G. YOGORE, JR., M.D.
Department of Parasitology, Institute of Hygiene
University of the Philippines

INTRODUCTION

The laboratory diagnosis of schistosomiasis japonica is, as a rule, not difficult if the examination is made during the dysenteric period when the ova are present in great numbers. The typical ovum of *Schistosoma japonicum*, because of its relatively large size and characteristic morphology, is easily recognized by competent laboratory workers.

However, when the disease is in the late chronic stage, laboratory confirmation is frequently very difficult, if not impossible. This is so because fibrotic changes that have occurred in the intestinal wall have imprisoned all or most of the eggs. Under such circumstance only a few eggs, if any, reach the intestinal lumen. It is precisely this stage in which a great many of the patients coming to the Philippine General Hospital and other City hospitals are seen. It is not strange, therefore, that the laboratory confirmation can seldom be given by routine laboratory procedures, even when the clinical picture leaves little room for doubt.

The demonstration of eggs in the feces is further made difficult by the action of chemotherapeutic drugs, notably fuadin or tartar emetic. During the administration of these drugs, the eggs cease to be viable and their number is reduced. It then becomes a very difficult matter to mark the exact time when no more eggs are passed out. Determination of this point during and after treatment is very important because it gives a good, although indirect indication, as to whether or not the liver still continues to receive the ova, which

* Read at the Section on Hygiene and Public Health, 40th Annual Convention of the Philippine Medical Association, held in Manila on May 9, 1947.

ultimately produce the cirrhotic changes frequently seen in the late stage.

Serological tests have been devised for laboratory diagnosis of the disease, but such tests as the intradermal, complement-fixation, and serum globulin reactions are not as valuable and specific as the direct demonstration of the ova. Moreover, although these tests may establish previous infection, they do not serve as a guide as to whether or not the adult parasites are still laying eggs or are already dead—two important considerations in the specific chemotherapy.

MATERIALS

The great majority of the 372 specimens in our series were from patients in the Philippine General Hospital; only a few were from the North General Hospital. All the specimens were examined on the same day, within a few hours after they were received. A few were stools that had stayed overnight in the hospital wards before being sent to us. As soon as a positive diagnosis was made in the laboratory for a particular patient, we made it a point to get the stools of the same patient daily or every other day thereafter.. We could do this, however, only with patients in the Philippine General Hospital for those of the North General Hospital were not easily accessible to us.

On receiving the stools, we noted and recorded their gross character. Each sample was thoroughly mixed, to make each portion examined representative of the whole. At least five ordinary coverslip saline preparations were made and the entire field was examined thoroughly for various ova and also for protozoan cysts and trophozoites, which, even if present, might not be demonstrable by the special technics used in this study. Two one-gram portions were then weighed: one for the acid-ether centrifugation and the other for the Faust-Meleney egg-hatching examination. Any excess specimen was weighed and another Faust-Meleney technic performed to parallel the one using 1 gm. of the sample.

TECHNICS

A. *The Acid-Ether Centrifugation.* Adapted from Weller and Dammin(5).

1. In 5 cc. of 40% HCl (40 cc. of concentrated HCl diluted to 100 cc. with distilled water) in a small vial, one gram of the stool

is stirred with a glass rod until thoroughly suspended (Fig. 1, Plate 1).

2. This suspension is filtered through one layer of moist gauze bandage (Mesh 36 x 43 sq. inch) or two layers of moist gauze stretched over a funnel and received in a 15 cc. centrifuge tube (Fig. 2, Plate I).

3. Then 5 cc. of ether is added to this filtrate, the tube closed with a gloved finger and vigorously shaken for one minute. (Fig. 3, Plate I).

4. The tube is centrifuged at around 1,500 r.p.m. for about two minutes (Fig. 4, Plate I).

5. The interphase film at the acid-ether junction is loosened by ringing with a clean wooden applicator, and the acid and ether layers are rapidly poured off and discarded.

6. All the centrifugate remaining in the tube is examined in several coverslip preparations, using normal saline for dilution when the preparation appeared too thick. All *Schistosoma japonicum* eggs seen were counted, and the other ova present were noted.

B. *The Faust-Meleney Egg-Hatching Technic.* Adapted from Faust and Meleney(2).

Two parallel examinations by this technic were performed for each specimen, using one-gram portion in one instance and any excess of the specimen, usually from 5-10 grams, in the other.

1. The weighed sample was stirred well by means of a glass rod in tap water in 300 cc. sedimentation glass (Fig. 1, Plate II) until the fecal material was uniformly dispersed. Then the mixture was allowed to stand until it settled.

2. When sedimentation had been more or less completed, which usually took from 20 to 30 minutes, the supernatant fluid was decanted, and the water was replaced and stirred again.

3. Sedimentations and decantations were repeated until the supernatant fluid became water clear or almost colorless.

4. After the last decantation, tap water was again added to the sediment and the mixture was transferred to a long-necked or an Erlenmeyer flask which was filled almost to the brim and allowed to stand overnight (Fig. 2, Plate II).

5. On the following morning, with the aid of a hand lens, the upper 2 or 3 centimeters of the level of water in the flask was examined for the free swimming miracidia that had hatched overnight (Figs. 3 and 4, Plate II) if the eggs of the *Schistosoma japonicum*

were present in the sample. An attempt was made to count the miracidia. This was possible only when the number was small; when it was too large, only a rough estimate was made. At times, the sediment in the bottom of the flask was also examined for dead miracidia, empty egg-shells or eggs that had not hatched.

In this series, the junior author performed the acid-ether centrifugation technic and the senior author did the Faust-Meleney technics for each specimen. In order to eliminate bias in favor of one or the other technic, positive results obtained by one of the authors were invariably verified by the other worker. In other words, one worker checked the results of the other in every case.

PRESENTATION OF RESULTS

Our series comprised a total of 372 specimens of 37 patients examined during the periods from December 6, 1946 to May 6, 1947 and from June 16, to July 31, 1947. Those received from May 7 to June 15 (Series No. 246-402) were excluded because the junior author was on leave and only the Faust-Meleney technics could be performed.

A preliminary report of this study was presented at the 40th Annual Convention of the Philippine Medical Association on May 8, 1947. The present report is the whole study including observations up to July 31, 1947. Each specimen was examined by the direct fecal smear by both authors; by acid-ether centrifugation by the junior author, and by the two parallel Faust-Meleney technics using 1 gram and excess of 1 gram, respectively, by the senior author. Only 24 out of 37 patients whose stools were repeatedly examined in our laboratory were confirmed as cases of schistosomiasis japonica. The results of these examinations done in the 372 specimens received by us are summarized in Table 1.

TABLE 1.—*Results of the Examination for Schistosoma japonicum of 372 Stool Specimens in this Series*

Technics Used	Number and Percentage of Positives		Number and Percentage of Negatives	
Direct Fecal Smear (D.F.S.)	109a	29.1%	263	70.9%
Acid-Ether Centrifugation Technic	140	37.6%	232b	62.4%
Faust-Meleney Egg-Hatching Technic (1 gram)	144	38.7%	228c	61.3%
Faust-Meleney Egg-Hatching Technic (5-10 grams)	170	45.7%	202d	54.3%
Combination of Methods	187	50.3%	185	49.7%

a Out of these 109 (+)'s by D.F.S., 12 turned out (—) by A-E.C.T.; 9 turned out (—) by F-M.E.-H.T. (using 1 gm. of stool); and 9 turned out (—) by F-M.E.-H.T. (using 5-10 gms. of stool).
b Out of these 228 (—)'s by A-E.C.T., 12 were (+) by D.F.S.
c Out of these 228 (—)'s by F-M.E-H.T. (using 1 gm.) 9 were (+) by D.F.S.
d Out of these 202 (—)'s by F-M.E-H.T. (using 5-10 gms.) 9 were (+) by D.F.S.

Table 1 shows that the number of positives found by direct fecal smear alone is definitely increased by the special technics under investigation. From 109 or 29.1% the number of positives rose to 140 or 37.6% by acid-ether centrifugation; to 144 or 38.7% by Faust-Meleney egg-hatching technic (using 1 gram); to 170 or 45.7% by Faust-Meleney egg-hatching technic (using 5-10 gms.); and to 187 or 50.3% by the combination of methods. This means that a combination of methods gave a maximum of 187 positives in a series of 372 samples giving a total increase of 78 positives by the combination of methods over the direct fecal smear alone.

Of interest were the results of the individual examination of each of the 78 negatives (by direct fecal smear) which turned out

positive by employing the special technics under study. These results are shown in Table 2 and summarized in Table 3.

TABLE 2.—*Individual Results of 78 Examinations Negative by D.F.S., but Positive by Any or All of the Special Technics under Study.*

Exam.	Serial No.	D.F.S.	A-E.C.T.	F-M.T. (1 gm.)	F-M.T. (5-10 gms.)
1	3	(—)	(—)	(—)	+
2	4	(—)	+	+	+
3	7	(—)	(—)	(—)	+
4	20	(—)	+	(—)	(—)
5	22	(—)	+	+	+
6	23	(—)	+	(—)	+
7	25	(—)	(—)	(—)	(—)
8	26	(—)	+	+	+
9	27	(—)	+	+	(-)
10	29	(—)	+	(—)	+
11	33c	(—)	(—)	(—)	+
12	34	(—)	(—)	(—)	(—)
13	35	(—)	+	(—)	+
14	36	(—)	+	(—)	(—)
15	37	(—)	+	(—)	+
16	38	(—)	(—)	(—)	+
17	40	(—)	+	+	+
19	50	(—)	(—)	+	+
18	43	(—)	+	(—)	(—)
20	52	(—)	+	(—)	(—)
21	55	(—)	+	+	+
22	59	(—)	+	(—)	+
23	62	(—)	+	+	+
24	67	(—)	+	(—)	+
25	71	(—)	(—)	(—)	+
26	74	(—)	(—)	+	+
27	79	(—)	(—)	(—)	+
28	89	(—)	(—)	+	+
29	134	(—)	(—)	(—)	+
30	135	(—)	(—)	+	+
31	139	(—)	+	+	+
32	140	(—)	+	(—)	+
33	153	(—)	+	+	+
34	155	(—)	(—)	+	+
35	157	(—)	+	+	+
36	160	(—)	+	+	+
37	166	(—)	+	+	+

TABLE 2 (continued)

Exam.	Serial No.	D.F.S.	A-E.C.T.	F-M.T. (1 gm.)	F-M.T. (5-10 gms.)
38	167	(—)	+	+	+
39	180	(—)	(—)	+	+
40	186	(—)	+	+	+
41	196	(—)	+	(—)	+
42	226	(—)	+	+	+
43	227	(—)	(—)	+	+
44	230	(—)	(—)	+	+
45	404	(—)	(—)	+	+
46	409	(—)	(—)	+	+
47	412	(—)	+	+	+
48	413	(—)	(—)	+	(—)
49	418	(—)	+	+	+
50	421	(—)	(—)	(—)	+
51	423	(—)	+	+	+
52	433	(—)	+	+	+
53	444	(—)	+	+	+
54	446	(—)	+	+	+
55	451	(—)	(—)	+	+
56	453	(—)	+	+	+
57	454	(—)	(—)	(—)	+
58	456	(—)	(—)	+	+
59	462	(—)	+	+	+
60	463	(—)	(—)	+	+
61	464	(—)	+	+	+
62	467	(—)	(—)	(—)	+
63	473	(—)	+	+	+
64	475	(—)	+	(—)	+
65	478	(—)	(—)	(—)	+
66	481	(—)	(—)	+	+
67	482	(—)	(—)	+	(—)
68	488	(—)	(—)	(—)	+
69	491	(—)	(—)	+	+
70	493	(—)	(—)	(—)	+
71	495	(—)	(—)	(—)	+
72	497	(—)	(—)	+	+
73	511	(—)	+	(—)	+
74	515	(—)	+	(—)	+
75	516	(—)	+	+	+
76	526	(—)	(—)	+	+
77	530	(—)	(—)	+	+
78	544	(—)	(—)	(—)	+

TABLE 3.—*Summary of Results in Table 2*

78 Examinations Negative by D.F.S., but Positive by Any or All of the Special Technics under Study

Technics Used	Number and Percentage of Positives		Number and Percentage of Negatives	
Direct Fecal Smear (D.F.S.)	0	0%	78	100.0%
Acid-Ether Centrifugation Technic	43	55.1%	35	44.9%
Faust-Meleney Egg-Hatching Technic (1 gram)	44	56.4%	34	43.6%
Faust-Meleney Egg-Hatching Technic (5-10 grams)	70	89.7%	8a	10.3%

a These 8 (—)'s by Faust-Meleney egg-hatching technic (using 5-10 grams) were found positive by the other two special technics. Please see Table 2.

For a better appreciation of the results presented in Table 2, it must first be pointed out that our series of 372 stool examinations was done upon a special request to examine for *Schistosoma japonicum* eggs. For this reason, the search for those ova was made painstakingly. The specimens in which eggs could not be found were examined for a longer time, using at least 5 cover-slip preparations, and even more, before they were finally considered negative. Moreover, the majority of the examinations were repeated almost daily for many consecutive days for each patient, whether positive or negative on first examination.

The selection, therefore, of these 78 negatives, out of 372 samples. all of which were later found to be positive, was quite rigid. As a matter of fact, there were 30 specimens that were negative by the different special technics which were found positive by the direct fecal smear (see footnote on Table 1). Positive results then, which which were obtained by the special technics being studied out of

these 78 rigidly selected negative specimens, should be credited to the corresponding technic employed.

Tables 2 and 3 show that out of the 78 rigidly selected negatives (by direct fecal smear), 43 (or 55.1%) turned out positive by acid-ether centrifugation; 44 (or 56.4%) became positive by the Faust-Meleney egg-hatching technic (using 1 gm.); and 70 (or 89.7%) became positive by the Faust-Meleney egg-hatching technic (using 5-10 gms.) It must be kept in mind that these 78 negatives by direct fecal smear were all subsequently found positive by any or by a combination of the technics used.

The maximum number of samples in this series of 372 specimens that were found positive reached 187. For the evaluation of the technics used in this study, we considered the positive results of 187 examinations as 100%.

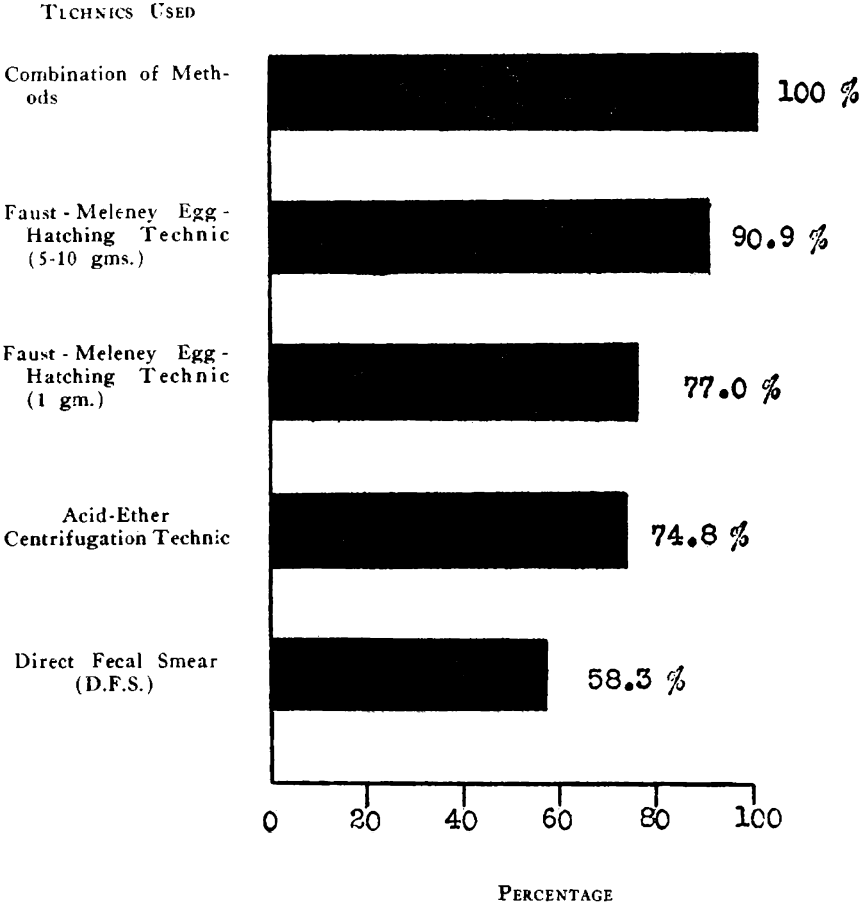
Our next problem was to find out whether or not the differences in the efficiency of the various technics that we tried were significant. Conscious of the rigid selection of our materials and the adequacy of the number of samples to be treated, we subjected the figures obtained to a statistical analysis. The results of this analysis are given in Table 4.

TABLE 4.—*Comparative Efficiency of the Different Technics Used in the Examination of 187 Positive Samples Measured Statistically* *

Technics Used	Number and Percentage of Positives		P = Probability That Their Difference Is Due to Chance
Direct Fecal Smear (D.F.S.)	109	58.3%	P = 0.0007
Acid-Ether Centrifugation Technic	140	74.8%	P = 0.6171
Faust-Meleney Egg-Hatching Technic (1 gram)	144	77.0%	P = 0.0004
Faust-Meleney Egg-Hatching Technic (5-10 grams) ..	170	90.9%	P = 0.0003
Combination of Methods ..	187	100.0%	

* We wish to thank Drs. P. Aragon and V. Valenzuela of the Department of Epidemiology and Bio-statistics, Institute of Hygiene, U. P. for helping us in the proper selection and in the statistical analysis of our data.

GRAPH 1.—*Comparative Efficiency in Percentage of the Different Technics Used in the Examination of 187 Positive Samples.*



The preceding table and graph definitely show that the difference between the efficiency of acid-ether centrifugation and that of direct fecal smear is significant. The first is superior to the second, the probability (p) that their difference was merely due to chance being 7 out of 10,000. The difference between the efficiency of Faust-Meleney egg-hatching technic (using 5-10 gms.) and that of the acid-ether centrifugation is also significant. The former is significantly better than the latter, the probability (p) that their difference was merely due to chance being 4 out of 10,000. The combination of methods, however, is significantly better than Faust-Meleney egg-hatching technic (using 5-10 gms.), the probability (p) that their difference was merely due to chance being only 3 out of 10,000

In contrast, the difference between the efficiency of acid-ether centrifugation and that of Faust-Meleney egg-hatching technic (using 1 gm.) was insignificant, the probability (p) that their difference was purely due to chance being 61 out of 100.

Based on this studies, we, therefore, consider the Faust-Meleney egg-hatching technic using 5-10 grams of the sample superior to all the other technics here studied. Our experience, however, gained during this investigation and borne out by statistical analysis of the results, has taught us that each of the technics studied has its own individual merits under particular circumstances, and, judging from the overall results, we feel justified in advocating the performance of the different technics, not excluding the direct fecal smear, in examining every fecal sample submitted from patients strongly suspicious for schistosomiasis japonica.

COMMENTS

In recent years, physicians and medical students in the Philippines have grown very "schistosoma-minded" and oftentimes a clinical diagnosis of schistosomiasis japonica is given to patients, especially with enlargement of both liver and spleen, whether or not they have had history of repeated attacks of dysentery, and whether or not they have come from known endemic areas. Many times we are unable to demonstrate the ova of *Schistosoma japonicum* even by repeated concentration or sedimentation methods, and the patient would be discharged with the final diagnosis incomplete or doubtful. This feeling of helplessness goaded us into searching for better methods of stool examination.

Just after the Liberation, we made a review(4) of the more recent, though scanty, literature available at the time, on new and tried methods that we might use in our laboratory. We encountered a report of Weller and Dammin(5), in which the acid-ether centrifugation and the zinc-sulphate centrifugation methods were tried in demonstrating the ova of *Schistosoma mansoni.* According to this study, the zinc sulphate centrifugation method was not satisfactory, but the acid-ether centrifugation was quite promising. We tried these two technics for *Schistosoma japonicum,* and we found the same results as those of the former workers.

This started our investigation on the subject, the results of which are reported in this paper. To simplify presentation, our present study was made entirely qualitative, as shown by our figures above. The quantitative study of our results is the subject of another paper. To these results, we may add a few more observations regarding our failures which were really instructive.

Thus, we have found that the character of the stool sample had much to do with such failures, especially in the acid-ether centrifugation, for stools that were rather hard and formed with only some mucous flecks on the surface were often negative by this method but positive by direct fecal smear. We believe that this is because the mucoid portions of the stools which invariably contain the eggs cannot be emulsified well even after stirring in the acid for a long time, and the eggs are not released from the mucous strands.

Moreover, mucus does not usually pass through the bandage gauze. If bits of mucus and eggs pass through, they usually float after centrifugation and become part of the interphase debris that is lost in the centrifugate. We have examined the material remaining in the gauze and the interphase debris, and we have succeeded in recovering the eggs which should otherwise be found in the centrifugate. Under this particular circumstance, it is advisable, therefore, to examine the mucous flecks found on the surface of such formed and hard stools by direct fecal smear first.

Many of our failures with the Faust-Meleney egg-hatching technic were, on the other hand, usually due to the presence of floating debris, which were usually pieces of mucus at the surface of the water which entangled and knocked out the miracidia causing their early disappearance. Examination of the sediment in such cases often revealed the presence of dead miracidia and empty egg-shells and occasionally a few unhatched immature or degenerated

eggs. This emphasizes the importance of seeing carefully that no floating debris is left in the flask overnight, and the need of checking on the sediment at the bottom of the flask whenever no miracidia are seen in the upper layer of the water.

In spite of these failures, however, the efficiency shown by the Faust-Meleney egg-hatching technic in this investigation remained high, although most of the stools received for examination came from patients who were at the time receiving fuadin—a factor that would operate adversely against its efficiency, if fuadin really renders eggs non-viable. This is perhaps because, in many of our positive samples found negative by all the other technics, one or two swimming miracidia seen by Faust-Meleney egg-hatching technic (using 5-10 gms.) sufficed to make the diagnosis and it is very much easier to spot 1 or 2 miracidia than to spot 1 or 2 ova in a sediment, whether obtained by acid-ether centrifugation or by other sedimentation methods. If the sample is negative by Faust-Meleney egg-hatching technic, therefore, sediment at the bottom of the flask should always be checked accurately during the course of fuadin treatment.

These are a few helpful suggestions in employing the Faust-Meleney egg-hatching technic. We recognize, however, that the real advantage of this technic lies in the unlimited amount of the fecal sample that can be used, multiplying thereby the chances of obtaining a positive result. The procedure is not complicated but it is rather tedious and time-consuming and may not be desirable as a routine procedure in a busy laboratory with a very limited personnel. Special investigations along this line, therefore, require additional trained personnel. Moreover, it is necessary to use fresh specimen and the results can be read only on the next day, or at most in the late afternoon of the same day, if it is started early.

In endemic areas, however, and in the provinces where the physician may not have a microscope, this technic would be useful; for he would need only a hand lens and some glassware, such as sedimentation glasses, Erlenmeyer flasks and glass or wooden stirrers and no chemical reagents. Physicians and medical students could, therefore, make use of this technic to advantage.

The acid-ether centrifugation technic, on the other hand, is particularly valuable when results are needed at once or when the stools are no longer fresh, for the procedure is fairly simple and less time-consuming. In fact, even formalinized stool specimens coming from

distant places and received by mail can be examined by this technic—a definite advantage to practising physicians who live far from established laboratories.

Another advantage of this technic is its high efficiency in recovering other helminth ova, including such operculated eggs as the Paragonimus ova. The limited amount of stool that can be examined, however, by this technic is one drawback; the less the amount of stool used, the less the chance of obtaining a positive result. It must be understood, moreover, that in laboratories that lack a centrifuge, this technic cannot be employed.

During the course of this investigation when we were almost halfway in our series we came across one publication by Weller and Dammin(6) and still later two more—one by Loughlin and Stoll(3) and another by Faust and Ingalls(1), both encountered in the March issue of the Tropical Disease Bulletin of this year describing certain improvements of the acid-ether centrifugation and sedimentation technics by the addition of wetting agents or detergents such as Triton NE, Duponol C, Naccomol NR, Tergitol Penetrant 08, Xyol, glycerine, etc. For lack of most of these chemicals at the time, and the difficulty of obtaining them, we could not start making parallel tests employing these modifications along with our own investigation. We proceded with our original project and it is to be hoped that further search for better technics would be continued until the best single laboratory method that will prove to be most reliable and most efficient is finally found.

SUMMARY AND CONCLUSIONS

1. A total of 372 fecal samples with special requests to search for *Schistosoma japonicum* ova were received by our laboratory from both the Philippine General Hospital and the North General Hospital from December 6, 1946 to May 6, 1947 and from June 16 to July 31, 1947.

2. Each specimen was examined by three methods: (1) the direct fecal smear, making at least 5 coverslip preparations; (2) the acid-ether centrifugation, using one gram of sample; and (3) the Faust-Meleney egg-hatching technic. The Faust-Meleney technic was performed in two parallel tests for each specimen, one examination using one gram of stool and another, if there was as excess, using 5 to 10 grams, depending on the excess amount available.

3. Out of 372 specimens, we found a maximum total of 187 that were positive for *Schistosoma japonicum* ova. With 187 positive examinations considered as 100% we made an evaluation of the efficiency of each technic and found the direct fecal smear to be 56.3% efficient, with 109 positive examinations; the acid-ether centrifugation 74.8% efficient, with 140 positive examinations; the Faust-Meleney technic (using one Gm.) 77% efficient, with 144 positive examinations; the Faust-Meleney technic (using 5-10 Gms.) 90.9% efficient, with 170 positive examinations; and the combination of methods 100% efficient, with the 187 samples taken as basis, being positive.

4. After subjecting the above figures to statistical analysis, we found: (a) that the acid-ether centrifugation was significantly superior to the direct fecal smear; (b) that the Faust-Meleney egg-hatching technic (using 5-10 Gms.) was significantly superior to the acid-ether centrifugation; and (c) that the combination of methods was significantly superior to any one single method, compared even with the Faust-Meleney egg-hatching technic using 5 to 10 grams. The difference between the acid-ether centrifugation and the Faust-Meleney egg-hatching technic (using 1 Gm.) was found to be not significant. This difference could be due to chance.

From the results of this study and the lessons learned during the investigations, we can recommend the Faust-Meleney egg-hatching technic (using 5-10 Gms.) as the more reliable single method of laboratory diagnosis for schistosomiasis japonica. If found negative, however, by this technic the process should further include additional examination of the sediment for dead miracidia, empty egg-shells and unhatched immature or degenerated egg. We strongly advocate, however, the performance of several different techniques together, not excluding the direct fecal smear.

REFERENCES

1. FAUST, E. C. and INGALLS, J. W.: The Diagnosis of Schistosomiasis japonica. III. Technics for the Recovery of the Eggs of *Schistosoma japonicum*, Amer. Jour. Trop. Med. 26: 559-583 (Sept.) 1946. (From Tropical Disease Bulletin, 44: 331-333 (March) 1947).

2. FAUST, E. C. and MELENEY, H.: Studies on Schistosomiasis japonica, Amer. Jour. Hyg. Monog. Series, No. 3, 1: 339 pp., 1923.

3. LOUGHLIN, E. H. and STOLL, N. R.: An Efficient Concentration Method (AEX) for Detecting Helminthic Ova in Feces (Modification of Telemann

Technic), Amer. Jour. Trop. Med. 26: 517-537 (July) 1946. (From Tropical Disease Bulletin, 44: 335-336 (March) 1947).

4. FESIGAN, T. P., REYES, F. A. and YOGORE, Jr., M. G.: Some Newer Knowledge in Parasitology: A Review, Jour. Phil. Med. Ass'n. 22: 189-104 (Sept.) 1946.
5. WELLER, T. H. and DAMMIN, G. J.: The Acid-Ether Centrifugation and the Zinc-sulphate Floatation Technics as Methods for the Recovery of the Eggs of *Schistosoma mansoni*, Amer. Jour. Trop. Med., 25: 167, 1945.
6. WELLER, T. H. and DAMMIN, G. J.: An Improved Method of Examination of Feces for the Diagnosis of Intestinal Schistosomiasis, Amer. Jour. of Clin. Path. 15: 11, 496-500 (Nov.) 1945.

PLATE I

Fig. 1. One gram of the stool sample is stirred with a glass rod in a small vial containing 5cc. of 40% HCl.

Fig. 2. The suspension is filtered thru moist gauze stretched over a funnel and received in a 15cc. centrifuge tube.

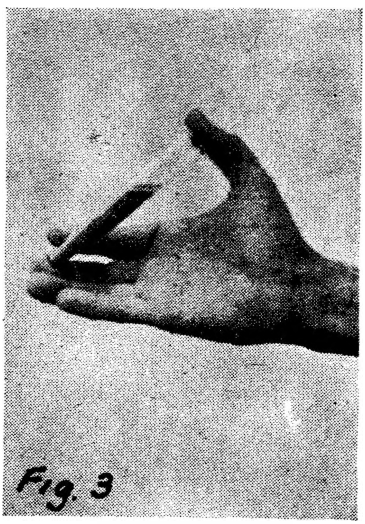


Fig. 3. Then 5cc. of ether is added to the filtrate, the tube closed with a gloved finger, vigorously shaken, and then centrifuged for 2 minutes.

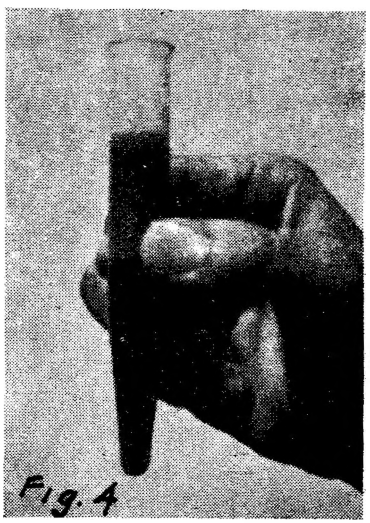


Fig. 4. The interphase film is loosened; the acid and ether layers poured off; and the centrifugate examined for *Schistosoma japonicum* eggs.

PLATE II

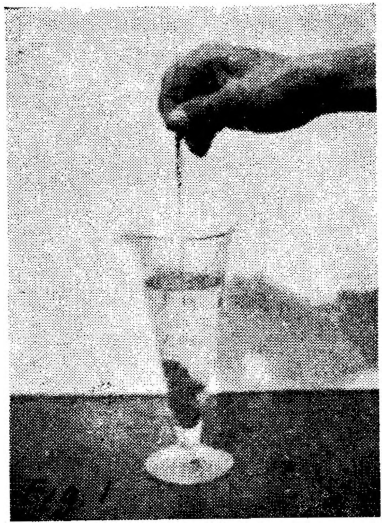


Fig. 1. The stool sample is stirred with a glass rod in a 300cc. sedimentation glass containing tap water and allowed to stand; the supernatant fluid is later decanted and the suspension is washed repeatedly.

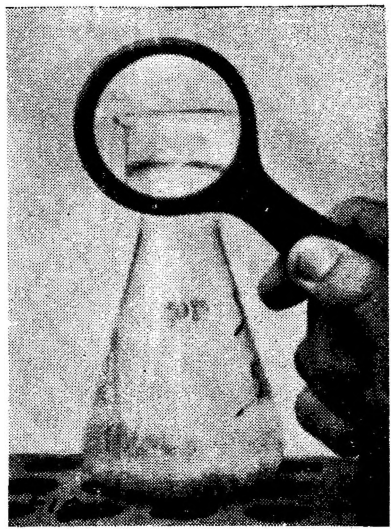


Fig. 2. After repeated washings and the supernatant fluid is water clear, the suspension is transferred to an Erlenmeyer flask, filled almost to the brim and allowed to stand overnight.

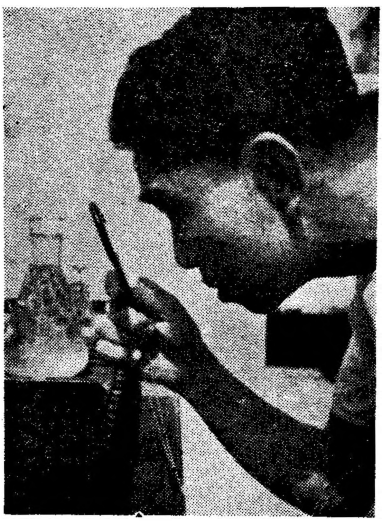


Fig. 3. Next morning, the upper level of the water in the flask is examined for miracidia with the aid of a hand lens.

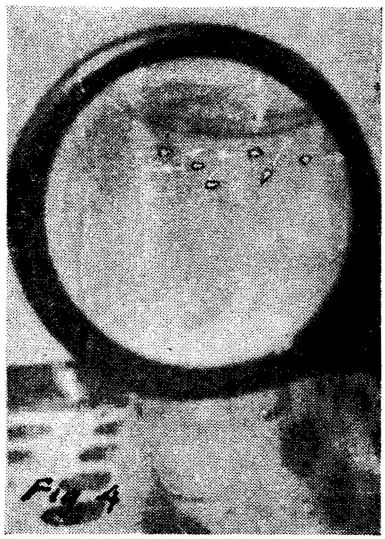


Fig. 4. The miracidia are small, minute, ciliated, ivory-white larvae; they swim actively in a more or less straight line.

Notice to Contributors

The ACTA MEDICA PHILIPPINA is devoted to the publication of original articles on medicine.

Manuscripts should be typewritten double- or (preferably) triple-spaced, and the original, not the carbon copy, sent to the Chief Editor, ACTA MEDICA PHILIPPINA. Articles are accepted only on condition that they have not been published elsewhere and that they will not be published previous to their appearance in the ACTA. References should conform to the following style:

UBALDO, A. R. and OCAMPO, G.: Incidence of Post Cataract Infection, Jour. Phil. Med. Assn. 18: 211-255 (April), 1938.

BEST, C. H. and TAYLOR, N. B.: The Physiologcial Basis of Medical Practice, 1939, The Williams and Wilkins Co., Baltimore, page 653.

Twenty-five reprints are supplied each author without charge. Additional copies may be had at the author's expense.

ACTA MEDICA PHILIPPINA

TABLE OF CONTENTS

The ACTA MEDICA PHILIPPINA is issued quarterly, one volume annually. The first volume began with the July-September, 1939 number. Subscription rates: Domestic—₱10.00; Foreign—₱12.00; Single copies—₱3.00.

Subscriptions are accepted for the full volume only. Remittances should be made payable to the University of the Philippines, Manila, Philippines.