

## STUDY REPORTS

1. Title : Clinical and Biochemical Studies of Beri—beri in Adults.

Principal Investigators: Vichai Tanphaichitr, M.D.  
Serene Vimokesant, D.Sc.  
Aree Valyasevi, M.D.

Assistant Investigator : Panga Viriyapanich, B.Sc.

Period of Report : 1 April 1967—31 March 1968

General Information Although the oldest Chinese medical book, Neiching, mentioned the disease beriberi almost 5000 years ago, it was not known for centuries that this illness was due to thiamine deficiency. In 1901, Grijns postulated that beriberi was caused by a lack of a protective factor found in the cortical part of rice and finally in 1936, Williams succeeded in synthesizing thiamine and determining its chemical formula. Diagnosis of beriberi is readily established in obvious cases, but a problem still exists in differentiating the subclinical form of this disease from other diseases. Many biochemical tests have been devised to solve this problem. However, no decision has been made re the best method to correlate clinical and subclinical manifestations of thiamine deficiency. Since beriberi is reported to be prevalent in Thailand, the present investigation was conducted in order to correlate clinical and biochemical findings in thiamine deficient patients.

Objectives

1. To describe the clinical manifestations of wet and dry beriberi patients.
2. To determine the level of urinary thiamine excretion and transketolase activity in normal subjects as compared to thiamine deficient patients.
3. To establish the relationship between urinary thiamine excretion and erythrocyte transketolase activity.
4. To correlate clinical manifestations of beriberi with the level of urinary thiamine excretion and erythrocyte transketolase activity before and after thiamine administration.

Description 1. Beriberi Patients. From August 1966 to November 1967 21 patients (22 observations, P1 was admitted twice, 10 months apart) were clinically diagnosed as beriberi and were studied at Siriraj Hospital. All but three patients were admitted to the medical ward and the other three (P10, P19, and P20) were studied as out—patients.

The diagnosis of beriberi is preliminary based on: 1) dietary history which is low in thiamine intake 2) edema with or without symptoms and signs of cardiac failure in wet beriberi 3) evidence of peripheral neuropathy without other known causes, and 4) good response to thiamine administration.

History and physical examination were recorded on a form specially designed for this study. Serial chest roentgenograms and electrocardiograms were performed before and after thiamine administration. Other laboratory examinations included routine blood and urine examinations. Determinations of the chemical composition of blood and cerebrospinal fluid were also carried out in cases when indicated. The levels of the erythrocyte transketolase activity (ETK Activity), thiamine pyrophosphate effect (TPP Effect) and urinary thiamine excretion were measured in these patients. Patients admitted to the hospital, blood and urine samples collected before they consumed hospital diets. Erythrocyte transketolase activity was measured by using Brin's technique (1) with some modification by Bunce and Sauberlich (2). The ETK activity is expressed as the disappearance of pentose in  $\mu\text{g}$  per gm hemoglobin of the hemolysate

per hour of incubation whereas TPP effect is expressed as per cent stimulation of the enzyme by the thiamine pyrophosphate added in vitro. The estimation of urinary thiamine excretion was done by thiochrome method (3) and urinary creatinine by using an auto analyzer (4). The urinary thiamine excretion is expressed as  $\mu\text{g}$  thiamine per gm creatinine.

All patients were initially treated with 100 mg thiamine hydrochloride given parenterally on the day of the admission after the initial collection of blood and urine samples and daily afterwards until the patients were markedly improved (1–8 weeks). This was followed by the oral administration for another 2 to 4 weeks. The ETK activity and TPP effect were again measured at 1 hour (30 minutes or 2 hours in some cases) and 24 hours after thiamine administration, whereas urinary thiamine was determined at 24 hours. During the recovery period, these two biochemical tests were performed occasionally. Blood and urine samples were collected before thiamine injection or in the morning before thiamine was given orally.

2. Control Subjects. Twenty-four Thai subjects (C1–C24), age ranging from 21 to 40 years, were included in this study. Among these subjects, three (C1, C2, and C3) are physicians. Physical examination, routine blood and urine tests, chest roentgenographic and electrocardiographic studies were performed and all subjects were found to be normal. Initial blood samples were drawn for the measurement of ETK activity and TPP effect. Urine was collected for the determination of urinary thiamine before breakfast. Only five subjects (C1, C2, C3, C9, and C22) received 100 mg of thiamine hydrochloride given parenterally after the collection of initial blood and urine samples. One and twenty-four hours after thiamine injection, blood samples were again drawn for the study of the ETK activity and TPP effect. Urinary thiamine was determined 24 hours after the vitamin injection.

3. Patients suffering from various diseases. ETK activity, TPP effect and urinary thiamine excretion were measured in 10 patients suffering from various other diseases (U1–U10). These two biochemical tests were performed in order to exclude the possibility of thiamine deficiency which might contribute to the presence of edema in patients U1 to U7 and peripheral neuritis in U8 to U10. These patients were treated with thiamine when first admitted to the hospital but no clinical improvement was observed. In all cases, the diagnosis was established after further investigations, for instance a large amount of arsenic was detected in the urine of patients U8, U9, and U10.

Progress      Clinical Description. Detailed descriptions of the patients are shown in Table 1. Fifty-four per cent of the patients were wet beriberi. It is of interest to note that 76.1% of the patients resided either in Bangkok or within 50 km from Bangkok and 62% of the patients were under 25 years of age.

Dietary History. All patients in this study consumed typical Thai diet which consists mainly of milled rice and small portions of cooked fishes and vegetables. Meat was occasionally consumed, except P7 and P8 who are siblings and gave the history that considerable amount of pork was consumed daily for 1 and 2 weeks prior to the hospital admission respectively. It is also of interest to note that patients P12 and P13 had changed from home-pounded glutinous rice to ordinary milled rice 2 and 1 months prior to the onset of disease respectively. In regard to the history of drinking alcohol, only P11 was a chronic alcoholic but had given up drinking for one year prior to the admission.

History of preceeding infection. Five patients had a history of preceeding infections about 1–2 weeks prior to the onset of the disease (P12, P15, P16, P17, and P21).

History of previous vitamin supplementation. Four patients (P17, P18, P20, and P21) had previously received vitamin injections before the admission.

The possible causative factors in these patients may be as follows:

1. Low thiamine intake. Previous studies showed that polished rice contained low thiamine (5) and protein. The thiamine intake was usually under 1 mg per day (6) and 50 to 85% of this vitamin was lost by customary methods of cooking (5).

Table 1. Description of Patients and Type of Beriberi

| Patients | Sex | Age    | Occupation | Residency      | Types of Beriberi |     |
|----------|-----|--------|------------|----------------|-------------------|-----|
| P 1      | OY  | Male   | 21         | Unemployed     | Nakornpathom      | Wet |
| P 2      | MB  | "      | 22         | Carpenter      | Hau-Hin           | "   |
| P 3      | PT  | "      | 60         | Unemployed     | Bangkok           | "   |
| P 4      | NC  | "      | 57         | Unemployed     | Bangkok           | "   |
| P 5      | RM  | "      | 43         | Carpenter      | Kanchanaburi      | "   |
| P 6      | TK  | "      | 18         | Student        | Samutprakarn      | "   |
| P 7      | ST  | Female | 17         | Factory worker | Bangkok           | "   |
| P 8      | SL  | "      | 15         | Factory worker | Bangkok           | "   |
| P 9      | PG  | Male   | 50         | Farmer         | Nontaburi         | "   |
| P10      | SC  | "      | 17         | Student        | Thonburi          | "   |
| P11      | AV  | "      | 48         | Unemployed     | Thonburi          | "   |
| P12      | HP  | "      | 17         | Blacksmith     | Bangkok           | Dry |
| P13      | KP  | "      | 17         | Jeweller       | Bangkok           | "   |
| P14      | PJ  | "      | 17         | Farmer         | Nontaburi         | "   |
| P15      | SP  | "      | 18         | Farmer         | Chacherngsao      | "   |
| P16      | KP  | Female | 16         | Trader         | Samutsakhorn      | "   |
| P17      | LT  | "      | 35         | Trader         | Thonburi          | "   |
| P18      | KT  | Male   | 51         | Shoemaker      | Bangkok           | "   |
| P19      | VB  | "      | 20         | Student        | Bangkok           | "   |
| P20      | LK  | "      | 66         | Farmer         | Suphanburi        | "   |
| P21      | SG  | "      | 15         | Watchmaker     | Nakorn Sawan      | "   |

2. Enzyme thiaminase. Even though most patients were young and physically active, it is unlikely that these factors were totally responsible for the etiology. Thiaminase has been found in fishes and some vegetables obtained from the North and Northeast of Thailand (7). The role the enzyme, thiaminase, plays in the etiology of beriberi in these patients is being further studied.

II. Clinical Manifestations In general, the clinical manifestations are not different from the previous reports (8, 9). Two major systems, the cardiovascular and nervous systems were involved:

1. The nervous system. Evidences of peripheral neuropathy were observed both in wet and dry types. Numbness as well as pain and tenderness over the calf muscles were the conspicuous findings. Superficial sensations (pain and touch) were more common than the joint position and vibration sensations. Motor weakness was detected in 12 out of 21 patients, showing difficulty in rising from the squatting position, whereas lame gait was observed in 9 out of 21 patients. It is observed that lower extremities were more affected than the upper extremities. Platt (10) postulated that it may be due to the length of nerves, amount of work done by the limbs and their blood supplies. Sixteen out of 21 patients had absent or hypoactive knee and ankle jerks. Alteration of tendon reflexes of upper extremities was less conspicuous. Except tendon reflexes, all of the mentioned abnormal findings disappeared within 4 months.

2. The cardiovascular system. The cardiovascular disturbances were only observed in wet beriberi. The outstanding changes following thiamine administrations were:

i. Diuresis was noted within 24 to 48 hours after the first dose of thiamine. The total body weight lost varied from 1.8 to 26.6 kg depending upon the severity of edema present (Table 2).

ii. Gallop rhythm and crepitation as noted in P1 and P11 also disappeared within 48 hours after thiamine administration.

Table 2. Changes in Body Weight before and after Thiamine Administration

| Patients | Before Treatment  |            | After Treatment         |                 |
|----------|-------------------|------------|-------------------------|-----------------|
|          | Body Weight (kg.) | B.W. (kg.) | Total Weight Loss (kg.) | Duration (days) |
| P 1 1st  | 67.0              | 50.0       | 17.0                    | 8               |
| 2nd      | 79.6              | 53.0       | 26.6                    | 13              |
| P 2      | 66.0              | 64.0       | 2.0                     | 3               |
| P 3      | 65.7              | 50.1       | 15.6                    | 13              |
| P 4      | 56.5              | 48.0       | 2.5                     | 7               |
| P 5      | 58.4              | 56.0       | 2.4                     | 3               |
| P 6      | 49.6              | 47.0       | 2.6                     | 3               |
| P 7      | 44.0              | 42.0       | 2.0                     | 6               |
| P 8      | 48.2              | 45.1       | 3.1                     | 4               |
| P 9      | 60.0              | 53.0       | 7.0                     | 8               |
| P10      | 47.8              | 46.0       | 1.8                     | 4               |
| P11      | 59.0              | 44.0       | 15.0                    | 13              |

Table 3. Changes in Blood Pressure and Heart Rate before and after Thiamine Administration

| Patients | Before Treatment |                 | After Treatment   |                       |                            |
|----------|------------------|-----------------|-------------------|-----------------------|----------------------------|
|          | B.P. mmHg.       | H.R. beats/min. | B.P. mmHg. 24 hr. | H.R. beats/min 24 hr. | B.P. mmHg. Recovery Period |
| P 1 1st  | 90/30            | 136             | 100/50            | 115                   | 120/80                     |
| 2nd      | 160/100          | 125             | 200/110           | 125                   | 120/80                     |
| P 2      | 120/70           | 93              | 120/70            | 79                    | 120/80                     |
| P 3      | 120/80           | 107             | 180/120           | 107                   | 150/100                    |
| P 4      | 160/90           | 115             | 170/90            | 60                    | 150/90                     |
| P 5      | 120/70           | 107             | 130/90            | 79                    | 130/90                     |
| P 6      | 120/60           | 71              | 120/70            | 60                    | 120/70                     |
| P 7      | 110/80           | 88              | 110/80            | 60                    | 110/80                     |
| P 8      | 110/80           | 83              | 110/80            | 62                    | 110/80                     |
| P 9      | 120/80           | 88              | 130/80            | 68                    | 130/80                     |
| P10      | 120/50           | 68              | 120/50            | 60                    | 120/50                     |
| P11      | 100/60           | 93              | 120/80            | 75                    | 140/80                     |

iii. Changes of heart rate and blood pressure are presented in Table 3. The mean heart rate was lowered significantly ( $P < 0.01$ ) within 24 hours after thiamine administration.

iii. Roentgenologic Findings of the Chest. Two significant roentgenologic findings, namely cardiomegaly and pulmonary congestion were frequently demonstrated in wet beriberi (Table 4). In 1942, Garland and McKenney reported that the enlarged heart shrank about 3 cm in its total transverse diameter in a period

Table 4. Roentgenologic Findings of the Chests in Wet Beriberi

| Patients | Cardiac Size                 |                              | D  | Lungs            |                 | D  |
|----------|------------------------------|------------------------------|----|------------------|-----------------|----|
|          | Before Treatment             | After Treatment              |    | Before Treatment | After Treatment |    |
| P 1 1st  | $\frac{15.0}{26.5}$ (56.60)* | $\frac{13.3}{26.0}$ (51.15.) | 10 | congestion       | clear           | 7  |
| 2nd      | $\frac{16.5}{26.6}$ (65.79)  | $\frac{11.9}{25.0}$ (47.60)  | 11 | congestion       | clear           | 7  |
| P 2      | $\frac{19.1}{27.0}$ (70.74)  | $\frac{13.0}{27.0}$ (48.15)  | 16 | congestion       | clear           | 6  |
| P 3      | $\frac{14.1}{26.5}$ (53.21)  | $\frac{13.5}{27.0}$ (50.00)  | 17 | congestion**     | clear           | 6  |
| P 4      | $\frac{14.8}{25.3}$ (58.50)  | $\frac{13.2}{24.6}$ (53.66)  | 18 | congestion       | clear           | 4  |
| P 5      | $\frac{14.6}{27.7}$ (52.71)  | $\frac{13.6}{26.5}$ (51.32)  | 8  | congestion       | clear           | 8  |
| P 6      | $\frac{10.9}{24.5}$ (44.49)  | $\frac{9.9}{24.7}$ (40.08)   | 14 | congestion       | clear           | 4  |
| P 7      | $\frac{12.9}{23.8}$ (54.20)  | $\frac{11.1}{23.7}$ (46.84)  | 11 | congestion       | clear           | 4  |
| P 8      | $\frac{12.0}{24.6}$ (48.78)  | $\frac{10.2}{23.1}$ (45.33)  | 15 | congestion       | clear           | 2  |
| P 9      | $\frac{11.0}{25.0}$ (44.00)  | $\frac{11.0}{25.0}$ (44.00)  | 7  | clear            | clear           | —  |
| P10      | $\frac{12.2}{24}$ (50.83)    | $\frac{10.9}{24}$ (45.41)    | 14 | clear            | clear           | —  |
| P11      | $\frac{16.2}{26}$ (62.30)    | $\frac{12.9}{26.9}$ (47.88)  | 29 | congestion**     | clear           | 14 |

\* Figures in brackets are per cent of cardiothoracic ratio.

\*\* Small amount of pleural effusion was present. D=Duration (days)

of 10 days of thiamine therapy (11). This was observed only in P1 and P2 (Fig. 1, 2). However, there was more or less decrease in cardiac size even in those who had the cardiothoracic ratio less than 50% after thiamine administration. From the present study it was shown that normal cardiothoracic ratio does not necessarily rule out cardiac involvement in patients with wet beriberi.

IV. Electrocardiographic Study. The electrocardiographic changes were found only in wet beriberi. The abnormalities present in these patients were:

1. 8 out of 11 patients showed the Q—Tc interval exceeding the normal values (0.42 sec. in men and 0.43 sec. in women). Their average Q—Tc interval was 0.45 sec.

2. Three patients (P1 during both admissions, P3 and P11) showed relative low voltages which increased after diuresis had begun. The low voltage was probably due to, partly if not totally, the presence of generalized edema.

3. Abnormalities of the T waves in the precordial leads were observed in 7 patients (P1 during both admissions, P2, P3, P4, P5, P6 and P11). This consisted of flat or inverted T waves (Fig. 3).

In those who attended the subsequent follow up, these abnormalities disappeared within 2 months after thiamine administration. The results on the electrocardiographic changes are in agreement with

CHEST ROENTGENOGRAM IN P2

BEFORE THIAMINE ADMINISTRATION



Fig. 1. The cardiothoracic ratio before thiamine administration in P2 is 19.1/27.0 (70.74%). Pulmonary congestion is also observed.

AFTER THIAMINE ADMINISTRATION

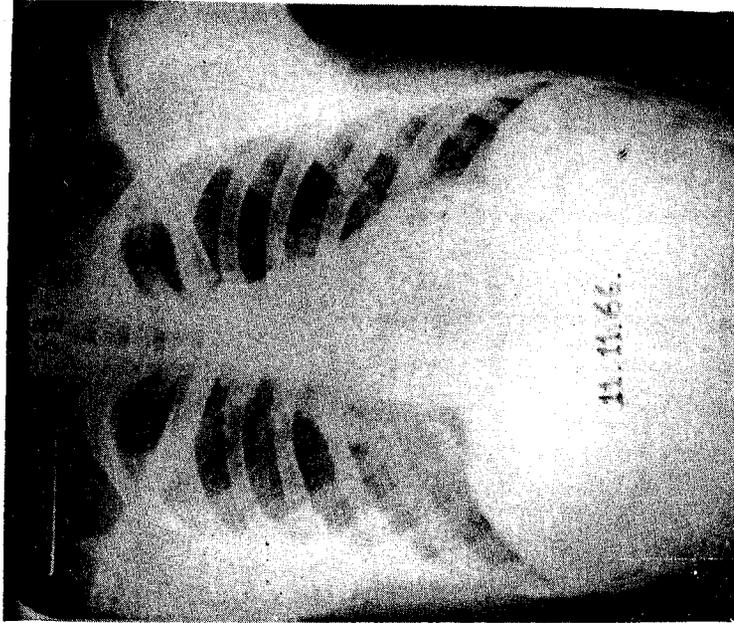


Fig. 2. Fifteen days after thiamine administration in P2, the cardiothoracic ratio is 13.0/27.0 (48.15%). Pulmonary congestion disappeared.

## ELECTROCARDIOGRAM IN P3

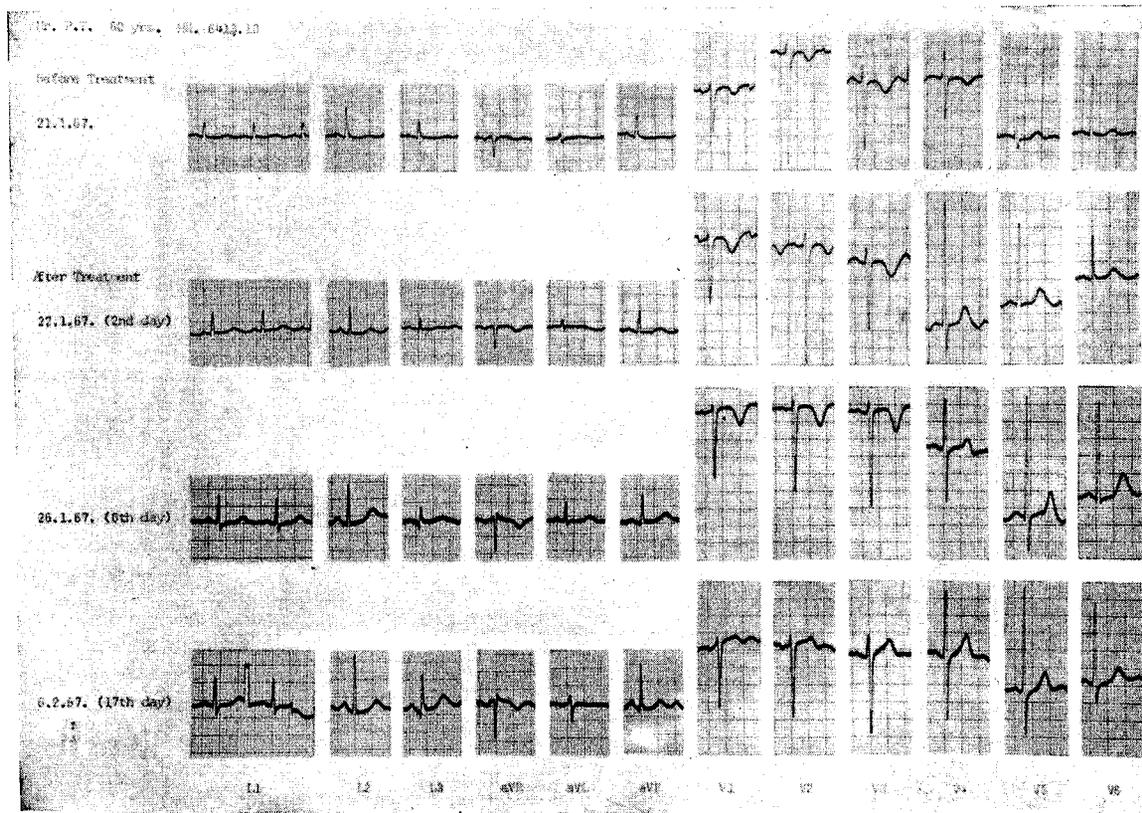


Fig. 3. In P3, before thiamine administration the Q-T<sub>c</sub> interval is 0.53 second and T waves are inverted in V<sub>1-4</sub>. During the recovery period T waves are more inverted in V<sub>1-3</sub>; 17 days after thiamine administration T waves are upright in V<sub>1-6</sub>. The Q-T<sub>c</sub> interval is 0.51 second.

Pallister (12), who showed no difference between Occidental and Oriental beriberi. Although the pattern of electrocardiographic changes is rather uniform it is by no means specific of the disease.

V. Anemia and Plasma Protein. The total white blood count, differential white count and morphology of red blood cells were normal. The detailed data of the levels of hemoglobin, hematocrit and plasma protein are presented in Table 5. Male patients of the present study showed significantly low mean hematocrit ( $P < 0.001$ ) whereas the mean hematocrit of female patients showed no significant difference ( $P = 0.16$ ) as compared to normal Thai Subjects reported previously (13). However, the number of female subjects is rather small. In the presence of normal morphology of red blood cells and absence of parasitic infestations in the present study, it is suggested that the anemia is not likely to be due to folic acid or vitamin B<sub>12</sub> deficiencies. However, mild iron deficiency anemia and thalassemia trait which are prevalent in Thailand (13, 14) could not be ruled out. The mean plasma protein also was significantly lower than the normal level reported previously. The results suggest that the patients were in the low or marginal protein nutritional status which may also have caused their anemia.

VI. Proteinuria and Changes in Blood Chemistry. Mild proteinuria (1<sup>+</sup>) was observed in P3 and P 11 before thiamine administration but this disappeared within 48 hours after thiamine administration. This might have been due to renal congestion.

Table 5. Levels of Hemoglobin, Hematocrit, Albumin, Globulin, and Total Plasma Protein in Beriberi Patients

| Patients |      | Hb<br>gm/100 ml | Ht<br>% | Albumin<br>gm/100 ml | Globulin<br>gm/100 ml | Total Protein<br>gm/100 ml |
|----------|------|-----------------|---------|----------------------|-----------------------|----------------------------|
| P 1      | 1st  | 8.4             | 30      | 2.9                  | 1.7                   | 4.6                        |
|          | 2 nd | 9.4             | 32      | 2.9                  | 2.0                   | 4.9                        |
| P 2      |      | 13.0            | 39      | 3.9                  | 2.6                   | 6.5                        |
| P 3      |      | 12.0            | 38      | 3.6                  | 2.2                   | 5.8                        |
| P 4      |      | 14.0            | 40      | 4.1                  | 2.5                   | 6.6                        |
| P 5      |      | 13.6            | 40      | 3.6                  | 2.5                   | 6.1                        |
| P 6      |      | 15.4            | 40      | 4.2                  | 3.8                   | 8.0                        |
| P 7      |      | 13.4            | 35      | 4.2                  | 3.3                   | 7.5                        |
| P 8      |      | 11.4            | 33      | 4.4                  | 2.2                   | 6.6                        |
| P 9      |      | 12.0            | 38      | 3.6                  | 1.7                   | 5.3                        |
| P10      |      | 12.3            | 38      | 5.3                  | 2.9                   | 8.2                        |
| P11      |      | 14.6            | 42      | 4.2                  | 3.8                   | 8.0                        |
| P12      |      | 12.0            | 40      | 3.7                  | 3.2                   | 6.9                        |
| P13      |      | 12.0            | 40      | 4.2                  | 3.8                   | 8.0                        |
| P14      |      | 15.6            | 43      | 4.6                  | 3.1                   | 7.7                        |
| P15      |      | 11.6            | 40      | 4.6                  | 2.4                   | 7.0                        |
| P16      |      | 12.2            | 40      | 3.9                  | 3.4                   | 7.3                        |
| P17      |      | 12.0            | 40      | 4.8                  | 3.4                   | 8.2                        |
| P18      |      | 15.5            | 47      | 4.2                  | 3.0                   | 7.2                        |
| P19      |      | 15.4            | 42      | 4.6                  | 3.7                   | 8.3                        |
| P20      |      | 9.4             | 30      | 4.1                  | 3.7                   | 8.8                        |
| P21      |      | 13.8            | 39      | 4.4                  | 2.4                   | 6.8                        |

Hematocrit (Mean±SD) Patients: male 38.78±4.31, female 37.00±3.55

Normal subjects (Na-Nakorn et al. 1966) male 45.96±3.13

female 40.20±2.67

Total Plasma protein (Mean±SD) Patients 7.01±1.15

Normal subjects (Vachanonda et al. 1964), Mean total plasma protein=7.49

Except P1 (first admission) and P11, all patients showed normal liver function, NPN, creatinine, BUN, and electrolytes. Elevation of NPN and creatinine without having pyrexia in P1 (NPN = 52 and creatinine = 2.4 mg per 100 ml) and P11 (NPN = 68.5 and creatinine = 2.7 mg per 100 ml) before treatment and became normal after thiamine administration (NPN = 30 and creatinine = 1.2 mg per 100 ml in P1; NPN = 46, creatinine = 1.6 mg per 100 ml in P11) may indicate the alteration of renal blood flow and glomerular filtration rate. The increase in alkaline phosphatase in P1 (before treatment = 9.04 B.U., 48 hours after treatment = 1.75 B.U.) and SGOT (before treatment = over 700 Sigma units; 36 days after treatment = 8 Sigma units) and SGPT (before treatment = 350 Sigma units; 36 days after treatment = 20 Sigma units) in P11 before thiamine administration probably indicates acute hepatic congestion (15).

VII. Findings of Cerebrospinal Fluid. Lumbar puncture was performed in 8 patients (P1, P2, P13, P14, P15, P16, P18, and P21). The cerebrospinal fluid was clear and colorless in all cases. The number of leucocytes was within normal limits. Protein content varied from 20 to 91 mg per 100 ml (mean ± S.E. = 47.62 ± 10.54). P12, P14, and P15 showed 91, 60, and 90 mg of protein per 100 ml respectively.

The significance of the increase in protein content in these patients could not be evaluated due to the lack of subsequent tapping during the recovery period.

### Biochemical Studies

The results of ETK activity, TPP effect and urinary thiamine excretion before and after thiamine administration within 24 hours in control subjects, previously non-treated beriberi patients, previously treated beriberi patients (those who received vitamin supplementation or enriched thiamine diet before admission to the hospital), and patients suffering from various diseases are presented in Tables 6, 7, 8A, 8B, 9 and 10 respectively.

I. The Interpretation of ETK Activity. The results show that no significant increase in the mean ETK activity was observed in control subjects at 1 hour after thiamine administration in vivo ( $P = 0.10$ ). Conversely, in non-treated beriberi patients, the mean ETK activity significantly increased both after the addition of thiamine pyrophosphate in erythrocyte hemolysate before thiamine administration and at 1 hour after thiamine administration in vivo ( $P < 0.001$ ). This may indicate that the transketolase coenzyme is not available during thiamine deficiency. This is confirmed by a significantly low ETK activity of non-treated beriberi patients when compared with that of control subjects ( $P = 0.02$ ). There was no significant difference between the mean ETK activity of the treated beriberi patients and that of the control subjects ( $P = 0.28$ ) (Table 11). The results of this study also show that the initial ETK activity in each non-treated beriberi was low, but at 1 hour after thiamine administration the ETK activity without the addition of thiamine pyrophosphate in vitro was equivalent to that observed when the initial blood sample was incubated with thiamine pyrophosphate. This was true in 10 patients whereas the 1 hour value was greater than the initial value in the other 6 patients (Tables 8A, 8B). The occurrence of the latter pattern could probably be explained by the reason that 1) the amount of thiamine pyrophosphate added in vitro ( $15.4 \mu\text{g}$  per 1 ml of initial assay tube volume) may not be sufficient to compensate for the amount depleted in vivo or 2) the mechanism of binding between the apotransketolase enzyme and thiamine pyrophosphate coenzyme in vitro is not as effective as in vivo. However, further investigation is necessary before any conclusion can be drawn. A significant increase in the ETK activity at 24 hours after thiamine administration when compared to the initial ETK activity was observed in the control subjects ( $P = 0.01$ ), in non-treated beriberi patients ( $P = 0.01$ ) and in treated beriberi patients ( $P = 0.015$ ). This increase may be due to a true increase in the ETK activity or to a natural variation. Dreyfus has stated that ETK activity may vary from subject to subject or from one time of the day to the other within the same subject (16). However, the results indicated that this could probably be explained by the latter reason, as is evidenced by the fluctuation of the level of the ETK activity from day to day (Fig. 4). However, the mean ETK activity at 2 hours and 24 hours after thiamine administration in the non-treated beriberi patients was not significantly higher than that of the 1 hour value after thiamine administration ( $P = 0.36$  and  $P = 0.46$  respectively).

II. The Interpretation of Thiamine Pyrophosphate Effect. The results of this study show that the mean TPP effect before thiamine administration in non-treated beriberi patient was significantly higher than that of the control subjects ( $P < 0.001$ ) whereas the mean TPP effect before thiamine administration in the treated beriberi patients and patients suffering from various diseases was not significantly higher than that of control subjects ( $P = 0.27$  and  $P = 0.52$  respectively) (Table 11). The mean TPP effect of the non-treated beriberi patients at 1 hour after thiamine administration was found to be reduced significantly from the initial value ( $P < 0.001$ )

When Sauberlich's criteria is used (17), the total number of subjects studied is classified as shown in Table 12. In the non-treated beriberi patients, except P19, they all exhibited TPP effect greater than 16%. All high per cent stimulation of the initial TPP effect was reduced to less than 10% within 1 hour after thiamine administration. (Tables 8A, 8B). P1 (second admission) and P5 exhibited TPP effect of less than 15% at 30 minutes after thiamine administration. Only P12, P15, and P16 showed TPP effect greater than 16% at 24 hours after thiamine administration (Fig. 5). This may indicate that although large amounts

Table 6. ETK Activity, TPP Effect and Urinary Thiamine Excretion in Control Subjects

| Subjects | ETK Activity disappearance of $\mu\text{g}$<br>Pentose/gm. Hb. of hemolysate/hr |          |            | TPP Effect<br>% | Urinary Thiamine<br>$\mu\text{g}/\text{gm}$ creatinine |
|----------|---|----------|------------|-----------------|--|
|          | No TPP*   | Plus TPP | Difference |                 |  |
| C 1      | 7,300   | 7,773    | 473        | 6.48            | 126  |
| C 2      | 6,970   | 7,421    | 451        | 6.47            | 84   |
| C 3      | 12,465  | 13,554   | 1,089      | 8.73            | 51   |
| C 4      | 5,152   | 5,152    | 0          | 0               | 18   |
| C 5      | 5,987   | 5,987    | 0          | 0               | 16   |
| C 6      | 6,288   | 6,288    | 0          | 0               | 40   |
| C 7      | 6,381   | 6,381    | 0          | 0               | 36   |
| C 8      | 6,919   | 6,919    | 0          | 0               | 79   |
| C 9      | 7,171   | 7,171    | 0          | 0               | 176  |
| C10      | 7,744   | 7,744    | 0          | 0               | 24   |
| C11      | 14,475  | 14,475   | 0          | 0               | 31   |
| C12      | 8,126   | 8,647    | 521        | 6.41            | 16   |
| C13      | 7,490   | 7,673    | 183        | 2.44            | 272  |
| C14      | 7,051   | 7,337    | 286        | 4.06            | 21   |
| C15      | 6,904   | 7,180    | 276        | 4.00            | 30   |
| C16      | 6,491   | 6,883    | 392        | 6.04            | 15   |
| C17      | 6,201   | 6,261    | 66         | 0.97            | 378  |
| C18      | 5,038   | 5,646    | 608        | 12.07           | 21   |
| C19      | 4,990   | 5,199    | 209        | 4.19            | 207  |
| C20      | 4,346   | 4,935    | 579        | 13.32           | 20   |
| C21      | 2,615   | 2,742    | 127        | 4.86            | 11   |
| C22      | 4,272   | 5,586    | 1,314      | 30.76           | 10   |
| C23      | 4,440   | 5,579    | 1,139      | 25.65           | 14   |
| C24      | 5,839   | 7,955    | 2,156      | 36.92           | 11   |

\* TPP=thiamine pyrophosphate

Table 7. Serial Changes in ETK Activity, TPP Effect and Urinary Thiamine Excretion in Control Subjects

| Subjects | Vit B <sub>1</sub> Injection |        | ETK Activity                                       |          |            | TPP Effect<br>% | Urine Thiamine<br>$\mu\text{g}/\text{gm}$ creatinine |
|----------|------------------------------|--------|--|----------|------------|-----------------|--|
|          | Before                       | After  | $\mu\text{g}$ Pentose/gm.Hb. of hemo./hr<br>No TPP | Plus TPP | Difference |                 |  |
| C 1      | Before                       |        | 7,300  | 7,773    | 473        | 6.48            | 126  |
|          |                              | 1 hr.  | 7,337  | 7,812    | 475        | 6.47            |  |
|          |                              | 24 hr. | 8,375  | 8,375    | 0          | 0               |  |
| C 2      | Before                       |        | 6,970  | 7,421    | 451        | 6.47            | 84   |
|          |                              | 1 hr.  | 7,355  | 7,401    | 46         | 0.62            |  |
|          |                              | 24 hr. | 7,575  | 7,803    | 228        | 3.10            |  |
| C 3      | Before                       |        | 12,465   | 13,554   | 1,089      | 8.73            | 51   |
|          |                              | 1 hr.  | 12,233   | 12,887   | 654        | 5.35            |  |
|          |                              | 24 hr. | 15,291   | 15,969   | 678        | 4.43            |  |
| C 9      | Before                       |        | 7,171  | 7,171    | 0          | 0               | 176  |
|          |                              | 1 hr.  | 7,553  | 7,745    | 192        | 2.54            |  |
|          |                              | 24 hr. | 7,744  | 8,260    | 462        | 5.97            |  |
| C22      | Before                       |        | 4,272  | 5,586    | 1,314      | 30.76           | 9  |
|          |                              | 1 hr.  | 5,279  | 5,905    | 626        | 11.86           |  |
|          |                              | 24 hr. | 5,259  | 5,259    | 0          | 0               |  |

Table 8 A. Serial Changes in ETK Activity, TPP Effect and Urinary Thiamine Excretion in Non-Treated Beriberi Patients P1-P6

| Patients             | Vit B <sub>1</sub> Injection |       | ETK Activity                              |          |            | TPP Effect<br>% | Urine Thiamine<br>μg/gm creatinine |
|----------------------|------------------------------|-------|---|----------|------------|-----------------|------------------------------------|
|                      | Before                       | After | μg Pentose/gm. Hb. of hemo./hr,<br>No TPP | Plus TPP | Difference |                 |                                    |
| P 1<br>1st admission | Before                       |       | 1,727                                     | 3,044    | 1,317      | 76.26           | —                                  |
|                      | 1 hr.                        |       | 5,150                                     | 5,412    | 262        | 5.09            |                                    |
|                      | 2 hr.                        |       | 3,105                                     | 3,398    | 293        | 9.44            |                                    |
|                      | 7 hr.                        |       | 3,210                                     | 3,210    | 0          | 0               | 85,941                             |
| P 1<br>2nd admission | Before                       |       | 6,008                                     | 8,695    | 2,687      | 44.72           | 22                                 |
|                      | 1 hr.                        |       | 7,694                                     | 8,349    | 655        | 8.51            |                                    |
|                      | 24 hr.                       |       | 7,591                                     | 9,327    | 736        | 9.70            | 157,757                            |
| P 2                  | Before                       |       | 678                                       | 1,257    | 579        | 85.40           | 110                                |
|                      | 1 hr.                        |       | 1,392                                     | 1,392    | 0          | 0               |                                    |
|                      | 2 hr.                        |       | 1,320                                     | 1,320    | 0          | 0               |                                    |
|                      | 24 hr.                       |       | 3,018                                     | 3,582    | 514        | 18.68           | 162,188                            |
| P 3                  | Before                       |       | 3,789                                     | 6,974    | 3,185      | 84.06           | 46                                 |
|                      | 1 hr.                        |       | 6,331                                     | 6,386    | 55         | 0.87            |                                    |
|                      | 2 hr.                        |       | 7,738                                     | 8,255    | 567        | 6.68            |                                    |
|                      | 24 hr.                       |       | 6,078                                     | 6,273    | 195        | 3.21            | 111,421                            |
| P 4                  | Before                       |       | 2,370                                     | 3,893    | 1,523      | 64.24           | 7                                  |
|                      | 1 hr.                        |       | 3,716                                     | 3,918    | 202        | 5.43            |                                    |
|                      | 2 hr.                        |       | 3,594                                     | 3,594    | 0          | 0               |                                    |
|                      | 24 hr.                       |       | 4,747                                     | 5,254    | 507        | 10.68           | 103,163                            |
| P 5                  | Before                       |       | 1,365                                     | 2,053    | 688        | 50.40           | 1.5                                |
|                      | 1 hr.                        |       | 2,722                                     | 2,722    | 0          | 0               | 0                                  |
|                      | 2 hr.                        |       | 2,727                                     | 2,727    | 0          | 0               |                                    |
|                      | 24 hr.                       |       | 2,464                                     | 2,464    | 0          | 0               | 31,466                             |
| P 6                  | Before                       |       | 4,206                                     | 5,852    | 1,646      | 39.13           | 1                                  |
|                      | 1 hr.                        |       | 5,603                                     | 5,786    | 183        | 3.27            |                                    |
|                      | 24 hr.                       |       | 6,507                                     | 7,005    | 498        | 7.65            | 3,836                              |

Table 8 B. Changes in ETK Activity, TPP Effect and Urinary Thiamine Excretion in Non-Treated Beriberi Patients P9—P16,P19

| Patients | Vit B <sub>1</sub> Injection |       | ETK Activity                |                               |                      | TPP Effect % | Urine Thiamine<br>μg/gm creatinine |
|----------|------------------------------|-------|-----------------------------|-------------------------------|----------------------|--------------|------------------------------------|
|          | Before                       | After | μg. Pentose/gm Hb of No TPP | μg. Pentose/gm Hb of Plus TPP | hemo/hour Difference |              |                                    |
| P 9      | Before                       |       | 3,213                       | 4,433                         | 1,211                | 38.01        | 8                                  |
|          |                              | 1 hr  | 7,463                       | 7,424                         | -39                  | 0            |                                    |
|          |                              | 24 hr | 4,357                       | 4,357                         | 0                    | 0            | 141,208                            |
| P10      | Before                       |       | 10,421                      | 12,521                        | 2,100                | 20.15        | 628                                |
|          |                              | 1 hr  | 12,598                      | 13,609                        | 1,011                | 8.03         |                                    |
|          |                              | 24 hr | 13,952                      | 14,028                        | 76                   | 0.54         | 814                                |
| P11      | Before                       |       | 9,140                       | 10,993                        | 1,853                | 20.27        | 23                                 |
|          |                              | 1 hr  | 11,427                      | 11,562                        | 135                  | 1.81         |                                    |
|          |                              | 24 hr | 10,772                      | 10,772                        | 0                    | 0            | 108,960                            |
| P12      | Before                       |       | 3,350                       | 4,020                         | 670                  | 20.00        | 636                                |
|          |                              | 1 hr  | 6,025                       | 6,340                         | 315                  | 5.23         |                                    |
|          |                              | 24 hr | 7,493                       | 8,351                         | 158                  | 11.45        | 83,145                             |
| P13      | Before                       |       | 2,148                       | 3,402                         | 1,254                | 58.38        | 0                                  |
|          |                              | 1 hr  | 5,972                       | 6,025                         | 53                   | 0.89         |                                    |
|          |                              | 24 hr | 5,670                       | 5,834                         | 164                  | 2.89         | 117,668                            |
| P14      | Before                       |       | 1,438                       | 2,605                         | 1,177                | 81.85        | 21                                 |
|          |                              | 1 hr  | 2,252                       | 2,302                         | 50                   | 2.22         |                                    |
|          |                              | 24 hr | 2,456                       | 2,456                         | 0                    | 0            | 91,811                             |
| P15      | Before                       |       | 4,532                       | 5,790                         | 1,258                | 27.76        | 583                                |
|          |                              | 1 hr  | 8,877                       | 8,877                         | 0                    | 0            |                                    |
|          |                              | 24 hr | 6,761                       | 8,727                         | 1,966                | 29.08        | 66,021                             |
| P16      | Before                       |       | 1,152                       | 1,368                         | 216                  | 18.75        | 45                                 |
|          |                              | 1 hr  | 2,520                       | 2,520                         | 0                    | 0            |                                    |
|          |                              | 24 hr | 2,592                       | 3,096                         | 504                  | 19.44        | 89,207                             |
| P19      | Before                       |       | 7,409                       | 8,337                         | 928                  | 12.53        | 37                                 |
|          |                              | 1 hr  | 8,610                       | 8,662                         | 52                   | 0.63         |                                    |
|          |                              | 24 hr | 9,478                       | 9,478                         | 0                    | 0            | 1,287                              |

Table 9. Serial Changes in ETK Activity, TPP Effect and Urinary Thiamine Excretion in Treated Beriberi Patients

| Patients | Vit B <sub>1</sub> Injection |        | ETK Activity<br>μg Pentose/gm. Hb. of hemo./hr |          |            | TPP Effect<br>% | Urine Thiamine<br>μg/gm creatinine |
|----------|------------------------------|--------|--|----------|------------|-----------------|------------------------------------|
|          | Before                       | After  | No TPP   | Plus TPP | Difference |                 |                                    |
| P 7      | Before                       |        | 9,104  | 9,206    | 102        | 1.12            | 35                                 |
|          |                              | 1 hr.  | 8,733  | 8,733    | 0          | 0               |                                    |
|          |                              | 24 hr. | 9,009  | 9,009    | 0          | 0               | 143,000                            |
| P 8      | Before                       |        | 10,047   | 10,047   | 0          | 0               | 48                                 |
|          |                              | 1 hr.  | 10,076   | 10,198   | 122        | 1.21            |                                    |
|          |                              | 24 hr. | 10,868   | 10,868   | 0          | 0               | 98,904                             |
| P17      | Before                       |        | 5,504  | 6,225    | 721        | 13.10           | 779                                |
|          |                              | 2 hr.  | 7,461  | 7,461    | 0          | 0               |                                    |
|          |                              | 24 hr. | 6,352  | 6,352    | 0          | 0               | 145,079                            |
| P18      | Before                       |        | 4,621  | 4,860    | 239        | 5.17            | 113                                |
|          |                              | 2 hr.  | 4,288  | 4,423    | 135        | 3.14            |                                    |
|          |                              | 24 hr. | 5,158  | 5,158    | 0          | 0               | 114,780                            |
| P20      | Before                       |        | 11,542   | 11,971   | 429        | 3.72            | 3                                  |
|          |                              | 1 hr.  | 12,411   | 12,411   | 0          | 0               |                                    |
|          |                              | 24 hr. | 12,556   | 12,556   | 0          | 0               | —                                  |
| P21      | Before                       |        | 12,145   | 12,782   | 637        | 5.24            | 39                                 |
|          |                              | 24 hr. | 14,135   | 14,425   | 290        | 2.05            | 3,409                              |

Table 10. ETK Activity, TPP Effect and Urinary Thiamine Excretion in Patients Suffering from Various Diseases before Thiamine Administration

| Patients | Diagnosis                 | ETK Activity<br>μg pentose/gm Hb of hemo/hour |          |            | TPP Effect<br>% | Urine Thiamine<br>μg/gm creatinine |
|----------|---------------------------|---|----------|------------|-----------------|------------------------------------|
|          |                           | No TPP  | Plus TPP | Difference |                 |                                    |
| U 1      | Iron deficiency<br>Anemia | 14,611  | 15,019   | 408        | 2.79            | 26                                 |
| U 2      | Iron deficiency<br>Anemia | 8,510   | 9,393    | 883        | 10.38           | 942                                |
| U 3      | Pellagra                  | 9,046   | 9,262    | 216        | 2.39            | 278                                |
| U 4      | Nephrotic syndrome        | 8,396   | 9,259    | 863        | 10.28           | 25                                 |
| U 5      | Cardiomyopathy            | 17,112  | 17,112   | 0          | 0               | 25                                 |
| U 6      | Cardiomyopathy            | 14,870  | 16,647   | 1,777      | 11.95           | 3,364                              |
| U 7      | Autoimmune disease        | 13,827  | 14,381   | 554        | 4.01            | 70                                 |
| U 8      | Arsenical neuropathy      | 12,871  | 12,871   | 0          | 0               | 329                                |
| U 9      | Arsenical neuropathy      | 13,843  | 14,711   | 868        | 6.27            | 4,606                              |
| U10      | Arsenical neuropathy      | 13,045  | 13,332   | 287        | 2.02            | 300                                |

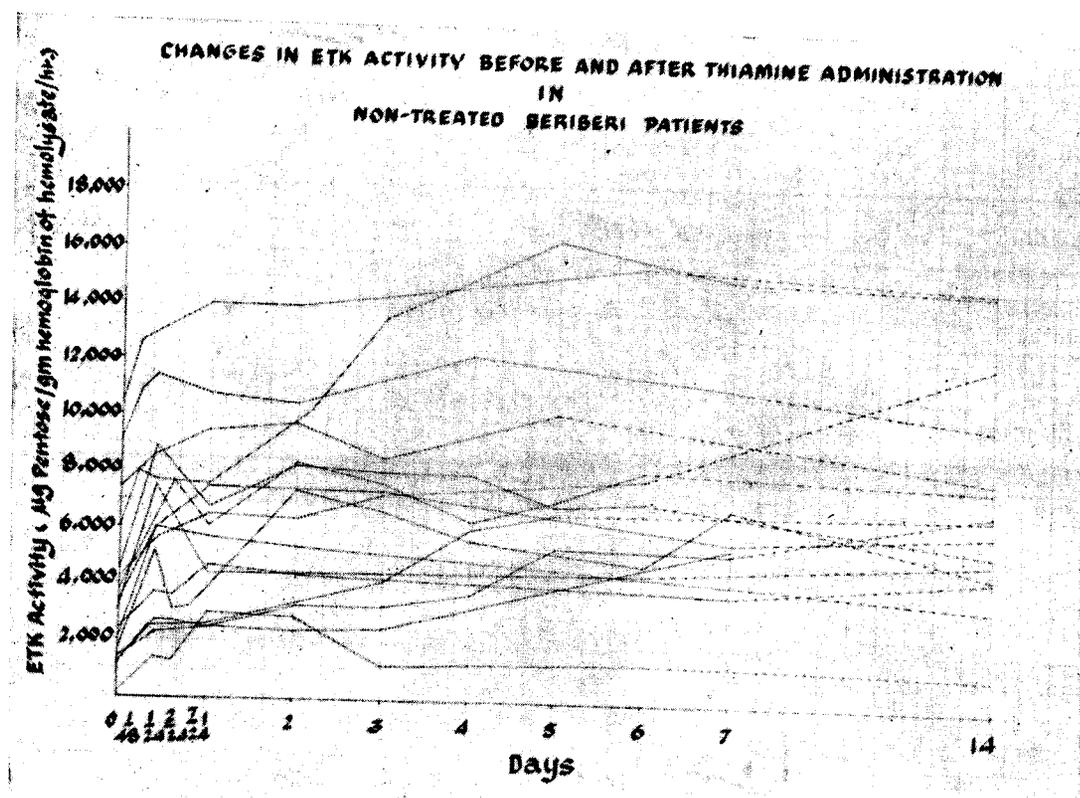


Fig. 4 Demonstrating erythrocyte transketolase activity. It can be seen that the ETK activity varies from one patient to another and from day to day even in the same subject. The initial ETK activity was consistently lower than the activity after thiaministration.

Table 11. Mean ETK Activity, TPP Effect, and Urinary Thiamine Excretion in Various Groups

| Subjects             | Number | ETK Activity* | TPP Effect** | Urine Thiamine*** |
|----------------------|--------|---------------|--------------|-------------------|
| Control              | 24     | 6,694±442#    | 7.22±2.07    | 71±20             |
| Non-Treated Beriberi | 16     | 3,934±724     | 46.31±644    | 145±63            |
| Treated Beriberi     | 6      | 8,827±1,273   | 4.73±1.89    | 170±123           |
| Various Diseases     | 10     | 12,613±1,163  | 5.01±1.41    | 997±514           |

\* Erythrocyte transketolase activity without addition of thiamine pyrophosphate in vitro, expressed as the disappearance of micrograms of pentose Per gram hemoglobin of the hemolysate per hour of incubation.

\*\* Thiamine pyrophosphate effect expressed as per cent stimulation of the enzyme in the presence of added thiamine pyrophosphate.

\*\*\* Urinary thiamine excretion expressed as micrograms of thiamine per gram creatinine.

# Values are Mean±SE.

n =number of patients for the calculation of mean urinary thiamine excretion.



of thiamine were given, the body could not retain thiamine very well. TPP effect also varied from day to day in each patient. However, the variation occurred fall below 13% stimulation during thiamine administration. From the mentioned findings it may be possible to conclude that TPP effect can be specifically used for judging thiamine adequacy.

III. Correlation between ETK Activity and TPP Effect. In 1965, Brin et al. (18) and Dibble et al. (19) reported that a reduced ETK activity was not always necessarily correlated to a high per cent TPP effect. This was also observed in the present study (Fig. 6).

IV. The Interpretation of Urinary Thiamine Excretion. Although the measurement of urinary thiamine excretion is a useful criteria for a survey study in a large population group, it is still subject to certain errors when applied to individuals as is evidenced by the results of this study. The mean urinary thiamine excretion of the non-treated beriberi patients was not significantly lower than that of the control subjects ( $P = 0.06$ ) (Table 11). This is not surprising when comparing urinary thiamine excretion in various subjects using ICNND's criteria (3). Even in control subjects, 17 out of 24 excreted urinary thiamine in the low and deficient ranges (Table 13). This may be due to the fact that urinary thiamine excretion may be a result of a decreased amount of thiamine in the diet on the previous day or on the day when the measurement is made (9). In 1954, Suzawa (20) reported that urinary thiamine tended to increase in beriberi despite a low blood thiamine and the presence of clinical manifestation. Three out of 15 non-treated beriberi patients in this study also exhibited a high urinary thiamine excretion before thiamine administration. The mean urinary thiamine excretion in patients suffering from various diseases was significantly higher than that of the control subjects ( $P = 0.005$ , Table 11). This may be due to the increased metabolism leading to a consequent increase of thiamine utilization. This explanation is supported by the fact that the mean ETK activity before thiamine administration of this group was significantly higher than that of the control subjects ( $P < 0.001$ ). A large amount of urinary thiamine excretion observed in control subjects and beriberi patients after thiamine administration (Tables 7, 8A, 8B, 9) could indicate an overflow of thiamine after such a large dose of the vitamin administration.

V. Correlation between Urinary Thiamine Excretion and TPP Effect. There was no correlation between these two biochemical tests in this study (Fig. 7), for example, 35 out of 55 control subjects excreted thiamine in low and deficient ranges as classified by ICNND but only 13 subjects of the same group showed the TPP effect greater than 16% (Table 14).

#### Correlation between Clinical Manifestations and the Biochemical Studies

The results of this study showed that those who excreted low urinary thiamine did not always exhibit clinical manifestations. Conversely, symptoms and signs of beriberi could be detected in some who had high thiamine excretion. Concerning TPP effect, all the non-treated beriberi patients showed initial TPP effect greater than 16%, except P19, but those who received previous vitamin supplementation or enriched thiamine diet, the initial TPP effect was below 15% in spite of the presence of the clinical manifestation of beriberi. Hence, a previous vitamin supplementation or an enriched thiamine diet should be considered in the interpretation of TPP effect, since this effect could be reduced within 1 hour (or 30 minutes in some cases) after thiamine administration. The results also demonstrated that 3 out of 24 control subjects showed a TPP effect greater than 16% without any clinical manifestation of beriberi (Table 6). In the one subject (C22) who received thiamine the high TPP effect was reduced to normal within one hour after thiamine administration (Table 7). Therefore, it seems possible to conclude that TPP effect may be used as a test to detect the inadequacy of thiamine at the cellular level even before the appearance of clinical manifestations.

Summary The clinical diagnosis of adult beriberi can be readily established in a full-blown case based on the following criteria:

1. Dietary history which is low in thiamine intake or food possibly containing the enzyme thiaminase.
2. Evidences of peripheral neuropathy which is present both in wet and dry beriberi. The important

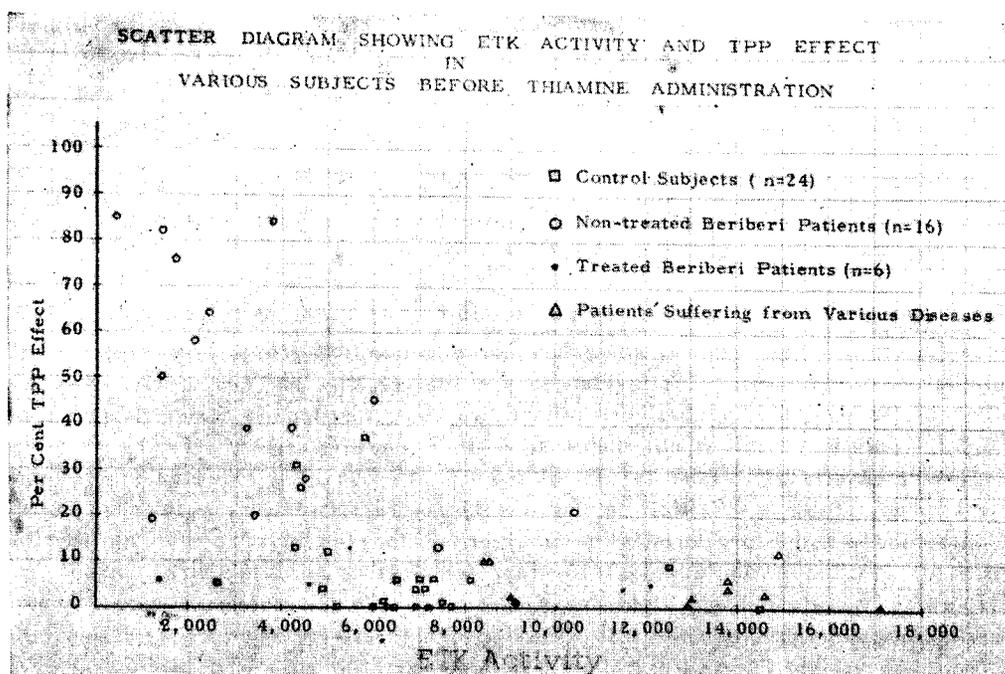


Fig. 6 Some negative correlation between ETK activity and TPP effect is observed when ETK activity is low. No such correlation was seen when ETK activity exceeded 6,000  $\mu\text{g. Pentose/gm. Hb.}$  of hemolysate/hr. TPP effect is expressed as per cent stimulation of the erythrocyte transketolase enzyme after the addition of  $2.61 \times 10^{-1} \mu\text{Mole}$  of thiamine pyrophosphate. ETK activity is expressed as the disappearance of  $\mu\text{g. pentose per gram hemoglobin}$  of the hemolysate per hour of incubation.

Table 13. Distribution of TPP Effect According to Urinary Thiamine Excretion in Various Groups

| Subjects                     | TPP Effect<br>% | Urinary Thiamine Excretion $\mu\text{g/gm creatinine}$ |          |           |
|------------------------------|-----------------|--|----------|-----------|
|                              |                 | $\geq 130$   | 66-129   | 27-65     |
| Control<br>n=24              | 0-15            | 4* (16.7)**  | 3 (12.5) | 6 (25.0)  |
|                              | 16-20           | —  | —        | —         |
|                              | > 20            | —  | —        | 3 (12.5)  |
| Non-Treated Beriberi<br>n=15 | 0-15            | —  | —        | 1 (6.7)   |
|                              | 16-20           | 1 (6.7)  | —        | 1 (6.7)   |
|                              | > 20            | 2 (13.2)   | 1 (6.7)  | 1 (6.7)   |
| Treated Beriberi<br>n=6      | 0-15            | 1 (16.7)   | 1 (16.7) | 3 (50.0)  |
|                              | 16-20           | —  | —        | —         |
|                              | > 20            | —  | —        | —         |
| Various diseases<br>n=10     | 0-15            | 6 (60.0)   | 1 (10.0) | —         |
|                              | 16-20           | —  | —        | —         |
|                              | > 20            | —  | —        | —         |
| Total n=55                   | 0-15            | 11 (20.0)  | 5 (9.1)  | 10 (18.2) |
|                              | 16-20           | 1 (1.8)  | —        | 1 (1.8)   |
|                              | > 20            | 2 (3.6)  | 1 (1.8)  | 1 (1.8)   |

Urinary Thiamine Excretion, ICNND's criteria; high  $\geq 130$ , acceptable 66-129, low 27-65 and deficient  $< 27$

TPP Effect, Sauberlich's criteria: acceptable 0-15, low 16-20, and deficient  $> 20$

\* Number of subjects; \*\* Figures in the brackets show % of the subjects in each group.

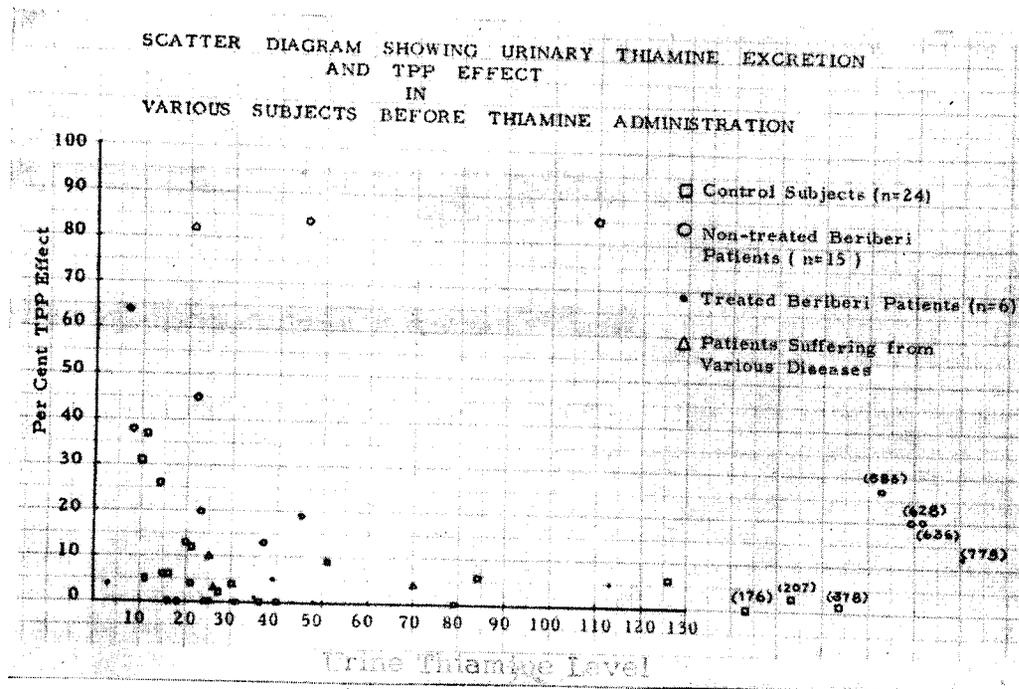


Fig. 7 No definite negative correlation is observed between TPP effect and urinary excretion. In some cases, the relationship is seen when urinary thiamine is low and the TPP effect is high.

Table 14 Urinary Thiamine Excretion in Various Groups of Subjects as Classified by ICNND's Criteria

| Groups               | Subjects |                 | Urinary Thiamine Excretion $\mu\text{g./gm. creatinine}$ |           |                  |  |
|----------------------|----------|-----------------|--|-----------|------------------|--|
|                      | Number   | High $\geq 130$ | Acceptable 66-129  | Low 27-65 | Deficient $< 27$ |  |
| Control              | 24       | 4 (16.70)*      | 3 (12.50)  | 6 (25.00) | 11 (45.80)       |  |
| Non-Treated Beriberi | 15       | 3 (20.00)       | 1 (6.67)   | 3 (20.00) | 8 (53.33)        |  |
| Treated Beriberi     | 6        | 1 (16.66)       | 1 (16.66)  | 4 (68.68) | —                |  |
| Various Diseases     | 10       | 6 (60.00)       | 1 (10.00)  | —         | 3 (30.00)        |  |

\* Figures in the brackets show per cent of the subjects in each group.

physical signs are the hypesthesia of pain and touch sensations, loss of ankle or knee jerks and difficulting in rising from squatting position including muscular tenderness at calf muscles.

3. Edema which is present in wet beriberi with or without the evidences of cardiac failure.

4. Patients respond to thiamine administration alone. The improvement appears faster in wet beriberi than in dry beriberi. The outstanding changes following thiamine administration in wet beriberi are:

a. Diuresis which appears within 24 to 48 hours after thiamine administration. The diuresis is evidenced by the reduction of initial body weight.

b. Heart rate may decrease and blood pressure may increase within 24 hours after thiamine administration.

c. More or less decrease in cardiac size as well as clearing of the pulmonary congestion is observed.

d. Abnormal electrocardiograms become normal.

Time taken for the recovery from motor weakness, abnormal sensations and reflexes requires months. Therefore it is very difficult to judge the immediate response to thiamine administration in dry beriberi; other possible known causes of peripheral neuritis must be carefully ruled out.

Concerning the biochemical aspects, from this study it may be concluded that 1) TPP effect is a good index for judging thiamine adequacy in human, even in the asymptomatic subjects. Up to the present time, the value greater than 16% can indicate a deficient state. However, this is only the arbitrary range and the previous vitamin supplementation and enriched thiamine diet must be considered in the interpretation of TPP effect. 2) The normal level of ETK activity can not be settled due to a great variation of this value even in control subjects, However, the increase of ETK activity from the initial low value together with a high per cent stimulation of TPP effect can be used to measure the thiamine adequacy in human. 3) The level of urinary thiamine excretion is less useful than TPP effect for diagnosing beriberi. However, it may be a useful criteria for a survey study in a large population group.