**CLXXXVIII. STUDIES ON THE METABOLISM
OF PYRUVIC ACID IN NORMAL AND
VITAMIN B₁-DEFICIENT STATES
V. THE EFFECT OF EXERCISE ON BLOOD PYRUVATE
IN VITAMIN B₁ DEFICIENCY IN MAN**

BY G. D. LU AND B. S. PLATT

From the Henry Lester Institute for Medical Research, Shanghai

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FOLLOWING the establishment [Platt & Lu, 1939] of the occurrence of increased amounts of pyruvic acid in the body fluids of patients suffering from beri-beri and the studies of Lu [1939] on the effect of muscular activity on the blood pyruvate of the rabbit, it appeared desirable that an investigation should be made of the relationships between pyruvic acid accumulation in the blood, amount of exercise and nutrition with respect to vitamin B₁; this has been carried out and the results are presented in this paper. There are a number of factors, of which increased muscular effort is one, which appear to contribute to the development of vitamin B₁ deficiency in man [Yang & Huang, 1934].

METHODS

The methods used for the estimation of pyruvic acid and bisulphite-binding substances (B.B.S.) have already been described [Platt & Lu, 1936; Lu, 1939].

Vitamin B₁ used for injection was made up (in a concentration of 1 mg./ml.) from the pure crystalline product in *N*/1000 HCl containing 20% by volume of ethyl alcohol [Kinnersley *et al.* 1933]. The exercise consisted either of (1) sitting up and lying down in bed without assistance 20 times, taking approximately 45 sec., or (2) climbing 100 steps five times, the test occupying 8–10 min., of which about half was occupied in climbing. Blood was taken for analysis (a) at rest, (b) immediately after exercise and (c) at the end of half an hour's rest after exercise.

RESULTS

Some of the results obtained from experiments on the effects of exercise on blood pyruvic acid and B.B.S. in cured and deficient cases are collected in the following tables.

It will be seen from the tabulated results in Tables I and II that the B.B.S. values give no consistent changes after slight exercise, while the pyruvate values of the deficient cases maintained a higher level after $\frac{1}{2}$ hr. as contrasted with the cured cases. This will appear more clearly from the detailed accounts of two typical examples given below.

Fig. 1 gives results on an acute type of beri-beri case with subacute manifestations of deficiency (oedema of legs and feet, dyspnoea and palpitation on exertion) for 2 months, worsening 2 days before admission to the fulminating form (described as + + + + by Platt & Lu [1936]). After taking blood for

Table I. *Cured cases*

Hospital no.	B.B.S. (expressed as pyruvic acid) mg./100 g. blood			Pyruvic acid mg./100 g. blood		
	Initial level	Immediately after exercise		Initial level	Immediately after exercise	
			$\frac{1}{2}$ hr. later			$\frac{1}{2}$ hr. later
1859	4.7	4.7	4.2	0.86	0.90	0.81
2115	3.4	4.8	3.5	0.40	0.60	0.53
2574	3.5	4.3	3.5	0.57	0.71	0.47
2910 ₁	5.9	6.7	3.2	0.02	1.13	0.53
2910 ₂	3.3	3.6	2.9	0.72	0.65	0.40
1936	2.6	3.3	2.3	0.40	1.01	0.58
2936	3.2	3.7	3.5	0.48	0.91	0.49
3127	2.9	3.7	2.8	0.54	0.76	0.45
3180	4.2	5.7	4.4	0.45	0.90	0.50
3198	3.5	3.8	—	0.63	0.84	0.61
3212	2.6	2.8	2.9	0.54	0.54	0.60
3212	3.7	3.2	2.6	0.79	0.87	0.71
3252	3.1	4.8	4.4	0.73	0.81	0.72
3319	3.0	3.4	3.0	0.40	0.50	0.40
3321	3.4	3.6	2.6	0.45	0.67	0.59

Table II. *Subacute cases—before and after treatment with vitamin B₁*

Hospital no.	B.B.S. (expressed as pyruvic acid) mg./100 g. blood			Pyruvic acid mg./100 g. blood		
	Initial level	Immediately after exercise		Initial level	Immediately after exercise	
			$\frac{1}{2}$ hr. later			$\frac{1}{2}$ hr. later
2707	2.6	3.4	4.30	0.59	0.74	0.88
2707	2.7	3.0	4.5	0.56	0.60	0.54
3012	2.6	3.1	3.4	0.71	1.02	1.00
3012	4.1	4.3	4.6	0.64	1.23	0.63
3146	3.9	5.4	5.4	0.94	1.10	1.47
3146	4.8	3.6	3.8	0.52	0.52	0.43
3146	3.6	3.5	4.4	0.66	0.68	1.52
3142	3.4	4.6	3.6	0.46	0.97	1.02
3142	3.1	4.2	3.7	0.91	1.05	0.58
3198	2.8	2.7	3.2	0.83	0.96	0.90
3198	2.7	3.0	3.5	1.50	0.52	0.60

Fig. 1. Case 3012. 1, Resting level; 2, immediately after exercise; 3, 30 min. later.

analyses, 10 mg. of crystalline vitamin B₁ were given intravenously. The patient recovered, the blood pyruvic acid level being restored to normal. He was put on to ordinary hospital diet, about 70 % of the calorie value of which was taken in the form of highly milled rice; the estimated intake of vitamin B₁ being about 0.5 mg. or less per day.

A tolerance test to exercise in bed was made on the 24th day after treatment with vitamin B₁. An increase of pyruvate after exercise was found to occur and to be maintained after half an hour. Two days later a similar amount of exercise was given 2 hr. after a dose of 2.5 mg. vitamin B₁. There was in this instance an increase of pyruvic acid immediately after exercise but the value returned to the original resting level at the end of a further half hour. After another period of 23 days on the vitamin-poor diet the test was repeated. There was a definite rise in blood pyruvate after exercise and the resting level was not restored at the end of half an hour. This is in contrast with the result of a repetition of the test 4 days later when 4 mg. vitamin B₁ were given 2 hr. before the exercise; there was a low resting level, and no rise of pyruvate either immediately or half an hour after exercise.

Subject no. 3198/36 had a history similar to the foregoing. A test of his response to exercise was made on the 12th day after treatment with 10 mg. of vitamin B₁. Only a slight increase of pyruvic acid in the blood was noted after exercise and the initial value was attained after the rest period. The "resting" level was found to be slightly raised on the 19th day under observation; an insignificant increase followed exercise in bed. It may be noted here that this type of response is commonly obtained in the subacute type of beri-beri and that after a dose of 4-5 mg. of vitamin B₁ it is usual to find a lowered resting value and little or no increase after exercise.

This patient, on his 42nd day in hospital was subjected to the stair-climbing test. The first set of 100 steps he completed in 45 sec., the second required 95 sec., and only 80 steps of the fourth set were climbed in 2 min. 47 sec. The subject complained of weakness of the legs, the pulse rate increased from a resting value of 70 beats per min. to 110; he was distressed, cyanosed and in general presented the appearance of a man in the fulminatory stage of beri-beri. The blood pressure before exercise was: systolic, 110, diastolic, 80 mm. After exercise: 10.30 a.m., 104/68; 10.40, 70/40; 10.45, 80/60; 11.40, 90/60 and 14.00, 95/50. The pulse rate fell to 88 per min. at 14.00 and then gradually fell to its original level in the course of the following 24 hr.

The pyruvic acid in the blood rose to nearly 2 mg./100 g. immediately after exercise and 15 min. later had reached a level which is characteristic of the cases of beri-beri of the most acute and "usually fatal" type described in the previous paper of this series. The second point of interest is that 1 hr. later the pyruvate was still present in the blood in considerably increased amounts; a normal value was reached some 4 hr. after the exercise. It will be seen from Table III that the B.B.S. increased to 11.6 in the acute deficiency state and to 8.4-9.9 mg./100 g. after exercise; these increases are far in excess of the amount accounted for by the accumulation of pyruvic acid (see Fig. 2).

380 ml. of urine obtained immediately before the exercise began contained 0.2 mg. pyruvic acid per 100 ml.: a specimen (390 ml.) collected at 14.15 had 1.76 mg./100 ml.

Similar exercise tests were carried out on two normal men (laboratory assistants) and three cured patients who received 2.5 mg. of vitamin B₁ 4 hr. before the test. The blood pyruvate value showed no appreciable increase in any of these subjects after exercise.

Table III. *B.B.S. and pyruvic acid*

	mg./100 g. blood	
	B.B.S.	Pyruvic acid
During B ₁ -hypovitaminosis		
On admission	11.6	3.8
1st day after B ₁ treatment	7.6	1.33
2nd day after B ₁ treatment	4.6	0.56
3rd day after B ₁ treatment	6.5	0.46
After severe exercise tolerance test		
Resting cured level	3.7	0.47
Immediately after severe exercise	8.4	1.86
$\frac{1}{2}$ hr. later	9.9	3.56
$1\frac{1}{2}$ hr. later	—	2.56
$4\frac{1}{2}$ hr. later	4.1	0.62

Fig. 2. Case 3198.

DISCUSSION

It has been shown in the previous paper of this series that when the urine of normal Chinese subjects contained increased amounts of "pyruvic acid" they had been engaged in some form of strenuous exercise. A similar observation had been reported by Johnson & Edwards [1937], who found that in normal subjects exercised to exhaustion pyruvic acid appears in the blood in amounts similar to those which are reached in severe beri-beri. Thus it is clear that it is possible to produce a state of exhaustion and a high level of pyruvic acid in the blood by an amount of muscular effort (stair-climbing) less than that which is well tolerated by subjects not deficient in vitamin B₁ and which can be accomplished without significant changes in the amount of pyruvic acid in the blood.

Another difference between the pyruvic acid change in the normal as compared with the vitamin B₁-deficient subject is in the time taken for the removal of the pyruvic acid which is produced during the exercise. In the experiments of Johnson & Edwards "normal" values were observed after less than 1 hr., whereas in the example described above, the high level of blood pyruvate is still maintained at the end of 1 hr. rest after the work is performed.

It may be mentioned here that the highest blood level is not immediately attained, no doubt for the reason that some time is taken for the metabolites to reach the blood stream from the cells in which they are formed. In B₁-deficient patients an amount of pyruvic acid greater than that found immediately after exercise has frequently been found in a test taken at the end of half an hour's rest following light exercise in bed. The need for time for products of

metabolism to reach the blood stream in B₁-hypovitaminosis may afford the basis of explanation for the fact that de Jong [1936] was unable to correlate the occurrence of clinical phenomena with changes in B.B.S. in the blood of vitamin B₁-deficient pigeons. It may also be noted that he did not find any increase of B.B.S. in his birds on "rousing" (exercising) them. An account of the blood pyruvate changes after exercise in normal rabbits has been given in a previous paper [Lu, 1939].

Results similar to those presented above were obtained for lactic acid by Inawashiro & Hayasaka [1928] for patients with beri-beri and for dogs with vitamin B₁ deficiency. They found that recovery was more prolonged in beri-beri than in the normal after exercise; that quite light exercise in the dog maintained a raised lactic acid level for long periods and that Na lactate injected into patients with beri-beri was synthesized to glycogen at much slower rates in the deficient as compared with the normal subject. Fisher [1931] also found that there was delayed removal of lactic acid from the heart and muscles of deficient pigeons after exercise. Lipschitz *et al.* [1938] have shown that polyneuritic birds are unable to remove injected pyruvate from the blood stream as rapidly as normal birds.

It has been demonstrated [Cowgill *et al.* 1931] that dogs which are exercised may be rendered deficient in vitamin B₁ in one-third to half the time required for those not exercised. Statements have been made to the effect that birds which have been rendered deficient have suddenly shown signs of the onset of the vitamin B₁-deficiency syndrome when movements are attempted on being set free. Ariyama [1931] has claimed to have shown by experiments on pigeons that vitamin B₁ can diminish exhaustion and promote recovery from fatigue. There is abundant evidence in the clinical literature on the aetiology of beri-beri [cf. Shimozona, 1931] that muscular effort may contribute to the onset of beri-beri and that in a given deficient subject a lesion may be related to the amount of work performed by a particular part of the body. It is probable that increased muscular work is only one of a number of factors belonging to a group which have in common the property of increasing the requirement for, or deficiency of, vitamin B₁ on account of their contribution to the increase of metabolic activities in which this food factor is involved. The view has been expressed [Platt, 1938] that, given vitamin B₁ deficiency, the clinical manifestations of the state may be determined more by the operation of stress factors than by differences in intake of vitamin B₁.

A practical issue of this investigation is the use which can be made of exercise to reveal latent vitamin B₁ deficiency states. The points which appear to be of most importance in designing a test are: (1) much less muscular effort is required to effect an appreciable increase of blood pyruvate in a vitamin B₁-deficient subject than in one normally nourished with vitamin B₁, (2) there is delayed removal of pyruvate from the blood after exercise during vitamin B₁ deficiency, and (3) the type of pyruvate response to exercise followed by dosage with vitamin B₁ also differs. Exercise tests may, however, be limited by the patient's inability to engage in heavy effort and it is sometimes undesirable that the patient be subjected to physical stress. In view of this it is suggested that a modification might be based on the determination of blood changes following a measured amount of work in a limb in which venous return is obstructed. In view of the possible association of fatigue, vitamin B₁ deficiency and exercise [Platt & Gin, 1934] this test might well be combined with observations of the development of sensations of pain in the exercised limb.

SUMMARY

1. Light muscular work by vitamin B₁-deficient human subjects is followed by an increase of blood pyruvate as measured by the 2:4-dinitrophenylhydrazone method. The rise of pyruvate may be maintained or still further increased at the end of half an hour's rest after the exercise.

2. After the intravenous administration of pure vitamin B₁, light exercise may or may not be followed by an immediate increase of blood pyruvate; following half an hour's rest after the exercise "resting levels" are found. This restoration of the level may be accounted for on the grounds that less pyruvate has been formed in exercise or that it has been more rapidly removed.

3. When the changes of blood pyruvate are inconsiderable, marked increase, accompanied by clinical manifestations resembling those of fulminating beri-beri, may follow heavier work. Still heavier work may, however, be well tolerated by subjects adequately supplied with vitamin B₁ and be insufficient appreciably to raise the blood pyruvate.

4. Values for blood pyruvate as high as those found in fulminating beri-beri may be attained after exercise of deficient subjects.

5. Similar high values have been observed by other workers to be reached by normal subjects when exercised to exhaustion. During rest after exercise, however, initial values are restored to normal subjects in less than 1 hr.

6. In vitamin B₁-deficient subjects there is delayed recovery after high values following exercise.

7. In an experiment in which high levels of blood pyruvate were produced in a deficient subject it was found that maximal values were not attained until some time after the completion of the exercise.

8. Bisulphite-binding substances (calculated as pyruvic acid) are increased when blood pyruvate is markedly increased, but to an extent greater than can be accounted for by the increase of pyruvic acid as estimated by the hydrazone method.

9. "Pyruvic acid" is excreted in increased amounts after exercise.

10. Evidences are recorded of circulatory changes accompanying accumulation of pyruvate after exercise of a deficient subject.

11. Excessive muscular work is discussed as one of a number of factors which may be responsible for the development of vitamin B₁ deficiency.

12. Exercise of the whole body combined with measurements of blood pyruvate may be used as a method for revealing latent vitamin B₁ deficiency.

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