The effect of exercise on the venous blood ammonium concentration in man

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SYNOPSIS Mild muscular exercise did not cause any significant rise in the ammonium concentration of venous blood draining the exercising forearm of control subjects or patients with cirrhosis. However, in both cirrhotic and non-cirrhotic subjects moderate exercise produced significant increases in venous blood ammonium values and these occurred earlier and were more prolonged in cirrhotic patients. Severe exercise caused larger increases in venous blood ammonium concentration in all subjects but there were no significant differences between the mean ammonium concentrations of the cirrhotic and control groups either before or after exercise. All ammonium values returned to their pre-exercise levels within half an hour of resting.

The exact mechanism of these phenomena is not fully understood but they are of practical importance in the study of blood ammonium metabolism in normal subjects and in patients with cirrhosis of the liver. The importance of making subjects rest for at least 30 minutes before obtaining blood for ammonium determination is emphasized in order to obviate misleadingly high readings due to muscular activity.

As long ago as 1925 Luck, Thacker, and Marrack found elevated venous blood ammonium levels in epileptic patients following grand mal seizures but could not ascribe this effect to muscular contractions because of their inability to show that vigorous muscular activity raised the blood ammonium level. In 1927 Parnas, Mozolowski, and Lewinskiw found that the ammonium concentration in venous blood obtained from the exercising forearm of normal subjects was increased and concluded that fatiguing exercise was responsible for this rise. These observations were later confirmed by Kalk and Boris (1933). In 1958 Schwartz, Lawrence, and Roberts showed that vigorous general exercise resulted in raised blood ammonium levels in normal subjects and that the increases in arterial ammonium levels caused by induced convulsions could be prevented by suppression of the muscular contractions. Bessman and Bessman (1955) and Bessman and Bradley

(1955) drew attention to the importance of peripheral tissues in ammonium metabolism. They showed that as the arterial ammonium concentration rises in patients with hepatic encephalopathy, the peripheral tissues take up correspondingly larger amounts of ammonium. These findings were later confirmed by Summer-skill, Davidson, Dible, Mallory, Sherlock, Turner, and Wolfe (1960) and by Webster and Gabuzda (1958), who also noted that in the terminal stages of liver disease the peripheral tissues may liberate ammonium into the systemic venous circulation.

In 1960 Allen and Conn showed that mild exercise resulted in an increase in the ammonium concentration of blood draining the exercising forearm of cirrhotic patients but not of normal subjects. More severe muscular exercise caused a significantly greater increase in the venous ammonium level in cirrhotic patients than in non-cirrhotic subjects. Severe fatiguing exercise resulted in large elevations which were approximately equal in cirrhotic and non-cirrhotic subjects.

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The present investigation was undertaken to study the effects of mild, moderate, and severe forearm exercise on venous blood ammonium concentrations of normal subjects and patients with cirrhosis.

Materials and Methods

Fifteen control subjects and 15 patients with cirrhosis took part in this investigation. All subjects were studied in the supine position after they had rested for a compulsory minimum period of 30 minutes. None of them had undertaken strenuous muscular exercise before the test nor were they permitted to clench their fists during the period of rest.

Forearm exercise entailed the use of a spring type hand-grip exerciser. Mild exercise consisted of 20 'power grips' at the rate of one per second. Forty 'power grips' constituted moderate exercise and 80 'power grips' severe exercise. Venous

blood samples were withdrawn from an antecubital vein through an indwelling heparinized needle immediately before starting forearm exercise and at intervals of 0, 1, 3, 6, 10, 15, and 30 minutes after exercise, care being taken each time to discard the first small portion of blood before collecting the sample for analysis. Puralin heparin, 500 IU in 2 ml normal saline, was used to prevent clotting in the indwelling needle.

Venous blood ammonium levels were estimated in all cases by the direct colorimetric method of McCullough (1967). In this method ammonia is determined using the indophenol reaction, the test being carried out on a deproteinized tungstate sulphuric acid extract of whole blood. The immediate treatment of blood samples by protein precipitant renders them stable for up to one hour, while subsequent centrifugation and separation of the supernatant permits storage in deep freeze for much longer periods before final analysis must be undertaken. The procedure is simple, rapid and specific, and is suitable for serial or multiple estimations.

Fig. 1 The effect of mild, moderate, and severe forearm exercise on the exercising forearm. The graph shows the mean venous ammonium concentration of groups of normal subjects and patients with cirrhosis A = control groups; B = cirrhotic groups; 1 = mild exercise; 2 = moderate exercise; 3 = severe exercise.

Results

The results are recorded in Table I and Fig. 1, all ammonium values being expressed as micrograms of ammonia nitrogen/100 ml blood.

MILD EXERCISE

Five control subjects and five patients with cirrhosis undertook this test. An analysis of variance showed no significant difference between the mean blood ammonium concentrations recorded before and at set intervals after completion of forearm exercise in either the control group or the cirrhotic group ($P > 0.05$ and $P > 0.05$ respectively). Mild forearm exercise did not therefore produce any significant elevation of the post-exercise blood ammonium concentration above the pre-exercise level in either control

subjects or patients with cirrhosis (Table II). Although there were significant differences between the ammonium concentrations of individual control subjects no such differences were noted between those of individual cirrhotic patients ($P < 0.01$ and $P > 0.05$ respectively). Comparison of the mean ammonium values of control subjects and cirrhotic patients showed that significantly higher values were recorded in the cirrhotic group both before and at set times after completion of forearm exercise but the differences were of the same order throughout the duration of the test (Table III).

MODERATE EXERCISE

The effect of 40 'power grips' was studied in five control subjects and five cirrhotic patients. An analysis of variance of the results obtained

Group	Type of Exercise	No. of Observations	Mean Venous Blood Ammonium Levels in $\mu\text{g}/100\text{ ml}$							
			Before Exercise	After Exercise						
				0 min	1 min	3 min	6 min	10 min	15 min	30 min
Control	Mild	5	63.2	79.2	74.8	80.0	72.6	74.2	71.4	63.2
Cirrhosis	Mild	5	139.8	128.8	137.4	136.8	138.4	127.6	133.8	120.4
Control	Moderate	5	75.6	97.8	123.8	103.0	88.4	80.4	80.2	76.2
Cirrhosis	Moderate	5	101.2	119.4	133.0	129.8	120.4	115.6	107.0	110.6
Control	Severe	5	93.2	170.8	177.8	177.6	161.2	135.2	118.4	98.4
Cirrhosis	Severe	5	104.6	151.4	156.8	154.8	142.8	117.2	118.6	112.0

Table I Mean venous blood ammonium levels in groups of control and cirrhotic subjects before and after mild, moderate, and severe forearm exercise

Group	Type of Exercise	Difference between Mean Pre- and Post-exercise Blood Ammonium Levels $\pm$ SE of Difference in $\mu\text{g}/100\text{ ml}$ at Different Times after Forearm Exercise							
		0 min	1 min	3 min	6 min	10 min	15 min	30 min	
Control	Mild	16.0 $\pm$ 9.3	11.6 $\pm$ 9.3	16.8 $\pm$ 9.3	5.4 $\pm$ 9.3	11.0 $\pm$ 9.3	8.0 $\pm$ 9.3	0 $\pm$ 9.3	
	Moderate	22.2 $\pm$ 14.7	48.2 $\pm$ 14.7 ¹	27.4 $\pm$ 14.7	12.8 $\pm$ 14.7	4.8 $\pm$ 14.7	4.6 $\pm$ 14.7	0.6 $\pm$ 14.7	
	Severe	77.6 $\pm$ 18.0 ¹	84.6 $\pm$ 18.0 ¹	84.4 $\pm$ 18.0 ¹	68.0 $\pm$ 18.0 ¹	42.0 $\pm$ 18.0 ¹	25.2 $\pm$ 18.0	5.2 $\pm$ 18.0	
Cirrhosis	Mild	-11.0 $\pm$ 18.1	-2.4 $\pm$ 18.1	-3.0 $\pm$ 18.1	-1.4 $\pm$ 18.1	-12.2 $\pm$ 18.1	-6.0 $\pm$ 18.1	-19.4 $\pm$ 18.1	
	Moderate	18.2 $\pm$ 7.5 ¹	31.8 $\pm$ 7.5 ¹	28.6 $\pm$ 7.5 ¹	19.2 $\pm$ 7.5 ¹	15.4 $\pm$ 7.5	5.8 $\pm$ 7.5	9.4 $\pm$ 7.5	
	Severe	47.2 $\pm$ 10.6 ¹	52.4 $\pm$ 10.6 ¹	50.4 $\pm$ 10.6 ¹	38.4 $\pm$ 10.6 ¹	12.6 $\pm$ 10.6	14.2 $\pm$ 10.6	7.6 $\pm$ 10.6	

Table II Comparison of pre- and post-exercise mean venous blood ammonium levels in groups of control subjects and cirrhotic patients

¹Significant at 0.05 probability

Type of Exercise	Cirrhosis minus Control Group Readings $\pm$ Standard Error of the Difference in $\mu\text{g}/100\text{ ml}$							
	Before Exercise	After Exercise						
		0 min	1 min	3 min	6 min	10 min	15 min	30 min
Mild	76.6 $\pm$ 19.0	49.6 $\pm$ 19.0	62.6 $\pm$ 19.0 ¹	56.8 $\pm$ 19.0 ¹	65.8 $\pm$ 19.0 ¹	53.4 $\pm$ 19.0 ¹	62.4 $\pm$ 19.0 ¹	57.4 $\pm$ 19.0 ¹
Moderate	25.6 $\pm$ 20.8	21.6 $\pm$ 20.8	9.2 $\pm$ 20.8	26.8 $\pm$ 20.8	32.0 $\pm$ 20.8	35.2 $\pm$ 20.8	26.8 $\pm$ 20.8	34.4 $\pm$ 20.8
Severe	11.4 $\pm$ 24.8	-19.4 $\pm$ 24.8	-21.0 $\pm$ 24.8	-22.8 $\pm$ 24.8	-18.4 $\pm$ 24.8	-18.0 $\pm$ 24.8	0.4 $\pm$ 24.8	13.6 $\pm$ 24.8

Table III Comparison of pre- and post-exercise mean venous blood ammonium levels in groups of control subjects and cirrhotic patients

¹Significant at 0.05 probability.

showed significant differences between the mean blood ammonium concentrations in both the control group and the cirrhotic group at the different times of sampling ($P < 0.05$ and $P < 0.01$ respectively). There was also a significant elevation of the mean venous blood ammonium concentration in the control group one minute after completion of exercise and in the cirrhotic group 0, one, three, six, and 10 minutes after completion of exercise (Table II). No significant differences were found between the mean venous ammonium levels of control subjects and cirrhotic patients either before or at the set times after moderate forearm exercise (Table III).

SEVERE EXERCISE

The effect of 80 'power grips' was studied in five control subjects and in five patients with cirrhosis. Analysis of variance of the results obtained showed significant differences between the mean blood ammonium concentrations in both the control group and the cirrhotic group at the different times of sampling ($P < 0.01$ and $P < 0.01$ respectively).

In the control group significant elevations of the mean venous blood ammonium levels above the pre-exercise levels were noted at 0, one, three, six, and 10 minutes after exercise and in the cirrhotic group at 0, one, three, and six minutes after exercise (Table II). Comparisons of the mean pre-exercise and post-exercise blood ammonium concentrations in the control group with those in the cirrhotic group did not show any significant differences (Table III).

No significant differences between the pre-exercise and 30-minute post-exercise venous blood ammonium levels were observed in either the control subjects or patients with cirrhosis, whether the exercise was mild, moderate, or severe (Table II). In other words the venous blood ammonium concentration in both control subjects and patients with cirrhosis returned to the pre-exercise level within 30 minutes of completion of all grades of forearm exercise.

Discussion

The cirrhotic patients taking part in this study had a higher resting venous blood ammonium level than the control subjects. Mild exercise did not cause any significant elevation of blood ammonium concentration in either group (Allen and Conn, 1960, found a small increase in their cirrhotic patients). The release of ammonium by moderately exercising muscle occurred in all subjects but began earlier and was more prolonged in cirrhotic patients than in control subjects. After severe exercise both cirrhotic and non-cirrhotic subjects exhibited equally large and prolonged releases of ammonium into the veins

draining the forearm muscles. Pilot studies showed that there were no simultaneous increases of ammonium concentration in the venous blood draining the contralateral resting limb.

The greater release of ammonium in cirrhotic patients after moderate exercise requires explanation. Allen and Conn (1960) postulated that as cirrhotic patients frequently have higher blood ammonium levels than non-cirrhotics they may, by virtue of continued uptake of ammonium by the peripheral tissues, accumulate ammonium in the muscles. The early release of ammonium observed in cirrhotics in the present investigation may simply reflect this saturation of voluntary muscles with ammonium. There is no evidence to indicate that the release of ammonium during exercise is related to a change in the pH gradient between intracellular and extracellular fluid, carbon dioxide tension, the degree of oxygen saturation, the increase in blood flow that accompanies muscular exercise, or the uptake of ammonium by the peripheral tissues (Allen and Conn, 1960).

Although the exact mechanism of ammonium release after muscular exercise is not fully understood, its occurrence emphasizes the need to rest subjects for at least half an hour before blood sampling in order to avoid misleadingly high readings. The phenomenon also indicates that the role of the peripheral tissues in ammonium metabolism must be carefully considered.

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