Biochemical aspects of motor neurone disease— Madras pattern

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SUMMARY A detailed study of four cases of MND-Madras has shown a significantly low plasma citrate level in these subjects. Comparison with other related groups of neurological disorders indicates the citrate/pyruvate ratio to be of diagnostic value in MND-Madras. This seems to suggest that the pathophysiology of MND-Madras may centre round altered citrate metabolism.

Motor neurone disease (MND), in general, is associated with an altered carbohydrate metabolism (Cumings, 1962; Shahani, Davies-Jones, and Russell, 1971). Motor neurone disease—Madras pattern (MND—Madras) (Meenakshisundaram, Jagannathan, and Ramamurthi, 1970), which has gained recognition (Spillane, 1972) as a distinct clinical entity, needs a basic biochemical orientation for comparative purposes.

METHODS

The subjects of the present study include 15 normal controls, four cases of MND—Madras, seven cases of other neuromuscular disorders, and four cases of diabetes mellitus. The 15 normal control subjects selected at random between the ages of 20 and 40 years are clinically and neurologically intact. The other groups are drawn from the inpatients of the Institute of Neurology, Madras, based on clinical, electromyographic (EMG), and histological diagnosis. Fasting blood samples were subjected to analysis in duplicate for various biochemical parameters like sugar, SGOT, SGPT, pyruvate, lactate, and citrate. Blood sugar was estimated by the method of Folin and Wu (1920), lactate following the method of Barker and Summerson (1941) and pyruvate adopting the procedure of Friedemann and Haugen (1943). The colorimetric method of Mohun and Cook (1957) was used for the assay of SGOT and SGPT. Citrate was estimated by the modification of Mascreen, Snehalatha, and Valmikinathan (1973) of the method of Beutler and Yeh (1959).

An oral glucose tolerance test (GTT) was carried out after 100 g of glucose load. Fasting, one hour and two hour samples of blood were subjected to biochemical analysis as indicated. All O.D. measurements were made with an ERMA photoelectric colorimeter model AE.11.

RESULTS

Levels of blood sugar, SGOT, SGPT, pyruvate, and lactate after glucose load in normal controls and subjects of MND—Madras are presented in Table 1. Subjects of MND—Madras show a greater increase in blood sugar level after one hour which does not come down to fasting level at two hours after the glucose load. There is a low but variable plasma lactate level and elevated pyruvate level which persists even at two hours after the glucose load. Other biochemical parameters such as SGOT and SGPT are within normal limits.

Fasting levels of amylase, sugar, pyruvate, and citrate in patients with MND—Madras are outlined in Table 2. The most significant finding is the critically low plasma citrate level together with an elevated pyruvate level.

Some biochemical investigations in normal controls and other study groups are detailed in Table 3 for comparison.

DISCUSSION

There seems to be an impairment in the utilization of glucose in MND—Madras as indicated

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TABLE 1
BIOCHEMICAL PARAMETERS AFTER GLUCOSE LOAD IN MOTOR NEURONE DISEASE (MADRAS PATTERN)

Particulars	(hr)	Sugar (mg/100 ml.)	Pyruvate (mg/100 ml.)	Lactate (mg/100 ml.)	SGOT (i.u./l.)	SGPT (i.u./l.)
Normal*	Fasting	80 ± 9.1	0.63 ± 0.12	8.8 ± 2.42	26 ± 3.10	31 ± 5.20
(4)	1	110 ± 11.3	0.72 ± 0.20	9.4 ± 3.84	25 ± 2.84	29 ± 4.60
	2	74 ± 3.1	0.68 ± 0.14	9.0 ± 4.02	26 ± 4.26	32 ± 6.30
Case 1	Fasting	88	1.9	2.5	20	31
R.R.	1	124	2.62	6.5	18	19
	2	94	2.28	4.8	14	40
Case 2	Fasting	88	1.36	4.75	47	16
S.K.	1	134	1.43	5.64	47	15
	2	106	1.36	5.25	44	15
Case 3	Fasting	80	2.5	6.25	15	32
T.D.	1	236	2.2	5.28	7	54
	2	156	2.0	7.2	9	11
Case 4	Fasting	96	1.83	7.4	9	6
B.K.	1	133	1.68	5.5	15	18
	2	112	1.64	5.0	17	21

* Mean plasma levels with standard deviation.

TABLE 2
PLASMA FASTING LEVELS OF SUGAR, PYRUVATE, AND CITRATE IN MOTOR NEURONE DISEASE (MADRAS PATTERN)

Case	Sugar (mg/100 ml.)	Amylase (Somogyi units)	Citrate (mg/100 ml.)	Pyruvate (mg/100 ml.)	Citrate Pyruvate
R.R.	88	136	1.10	1.90	0.63
S.K.	88	142	0.92	1.36	0.67
T.D.	80	106	0.80	2.5	0.30
B.K.	96	156	1.10	1.83	0.16

by lag GTT curves and elevated pyruvate level after oral glucose load (Table 1). This is similar to the pattern of biochemical changes reported in classical motor neurone disease (MND) under similar experimental conditions (Cumings, 1962; Shahani *et al.*, 1971).

The elevated plasma pyruvate level in classical MND has been attributed to many factors such as pancreatic dysfunction (Quick, 1969), spasticity (McArdle, Mackenzie, and Webster, 1960), and others. In our series of MND-Madras, the plasma amylase level is well within normal limits (Table 2), indicative of normal pancreatic function.

However, we have observed, in these subjects, that the fasting plasma pyruvate level itself is higher than in the normal controls and this elevated level persists even at two hours after an oral glucose load (Table 1). In this respect it differs from the classical MND where the pyruvate level is usually within normal limits and

tends to rise after glucose load (Shahani *et al.*, 1971). Further, there is generally a rapid increase in blood sugar level at one hour after glucose load (Table 1). These findings are suggestive of some involvement of certain nutritional factors as the underlying cause for the observed biochemical changes in these subjects.

The results so far discussed have clearly indicated an elevated pyruvate level in MND-Madras similar to classical MND. Since pyruvate plays an important role as a link between aerobic and anaerobic phase of carbohydrate metabolism, we extended our studies to some biochemical parameters beyond pyruvate in glucose oxidation. Such a study has been most rewarding in that we are able to show for the first time the critically low plasma citrate level in MND-Madras.

To assess the significance of this interesting finding, we have made a study of plasma citrate in neuromuscular disorders such as myotonia,

TABLE 3

COMPARATIVE VALUES OF SOME BIOCHEMICAL PARAMETERS IN NORMAL SUBJECTS AND OTHER GROUPS

Group		(no.)	Citrate (mg/100 ml.)	Pyruvate (mg/100 ml.)	Citrate Pyruvate
1	Normal	15	1.8 ± 0.7	0.7 ± 0.12	3.1 ± 0.53
2	Motor neurone disease*† (Madras pattern)	4	0.98 ± 0.14	1.89 ± 0.44	0.44 ± 0.72
3	Neuromuscular disorders	7	2.51 ± 0.36	0.56 ± 0.36	3.93 ± 1.36
4	Diabetes mellitus	4	3.13 ± 0.13	1.10 ± 0.07	2.57 ± 0.17

* P < 0.05 as compared with group 1.

† P < 0.01 as compared with group 3.

spinal muscular atrophy, etc, as muscle biopsy in MND—Madras has revealed a neurogenic atrophy (Figure). We have extended our citrate studies to diabetes mellitus where the pyruvate level is high. A moderate rise in plasma citrate in both the groups has been documented (Haerer, 1971; Mascree, 1973) (Table 3).

The low plasma citrate level in MND—Madras (Table 2) is highly significant statistically as compared with normal and other related study groups (Table 3). We have attempted to utilize the citrate/pyruvate ratio as an index of differentiation among metabolic disturbances associated with the defective utilization of carbohydrate. Such a ratio is greatly altered in MND—Madras (Table 3) and may possibly serve as a useful index for diagnosis.

Based on a careful study of four cases of MND—Madras, we are able to show for the first time that a biochemical lesion in MND—Madras relates to altered plasma citrate level. The low plasma citrate in MND—Madras may possibly be related to a slow turnover involving the condensing enzyme or to increased tissue uptake. Increased citrate uptake by tissue has been encountered in animals with muscular dystrophy (Tausky, Washington, Zubillaga, and Milhorat, 1962).

The results of the present study suggest that MND—Madras may well be a distinct entity biochemically as well as clinically (Meenakshisundaram *et al.*, 1970; Spillane, 1972) but as previous studies in classical MND have not made any reference to citrate, a study of plasma citrate level in classical MND is required to substantiate our concept.

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FIGURE Photomicrograph of transverse section of muscle showing classical grouped atrophy and small angulated fibres (arrows) indicating neurogenic atrophy. H and E section.

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