CCXLV. THE CHOLINE-ESTERASE CONTENT OF BLOOD IN MYASTHENIA GRAVIS

BY EDGAR STEDMAN AND WILLIAM RITCHIE RUSSELL

From the Department of Medical Chemistry, University of Edinburgh

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THE discovery by Walker [1934; 1935], amply confirmed by other investigators, that the symptoms of myasthenia gravis could be temporarily relieved by the parenteral administration of physostigmine or, with better results, of prostigmin, one of its synthetic analogues, has led to the suggestion [Hamill (see Walker, 1935); Pritchard, 1935] that the abnormal fatiguability in myasthenia is due either to a deficiency in the liberation of acetylcholine or to an abnormally rapid destruction of this substance. These suggestions were the logical outcome of the demonstration by Dale and his co-workers [e.g. Dale *et al.* 1936] that the transmission of an impulse from a motor nerve to its effector organ is brought about by the liberation from the nerve ending of acetylcholine. If the latter were released in subnormal amounts, muscular weakness would clearly result. The same effect could, however, conceivably be produced by an abnormally rapid destruction of the acetylcholine, liberated in adequate quantity, owing to the presence of excessively high concentrations of choline-esterase, the enzyme which destroys acetylcholine and hence prevents its accumulation in the body. In either case, the relief afforded by prostigmin would be due to its known inhibitory action on choline-esterase.

The present investigation was commenced some years ago in the hope that the data obtained by the estimation of the choline-esterase content of the blood of patients suffering from myasthenia gravis would permit us to distinguish between the above two possibilities. A statement of the broad results of the investigation, which has shown a low content of the enzyme in the serum, and an approximately normal one in the corpuscles, of such patients, has already been published by one of us [Stedman, 1935]. Since that time, however, Hicks & Mackay [1936] have claimed, as a result of the examination of one case of myasthenia gravis, that the disease is associated with a high content of choline-esterase in the serum, while McGeorge [1937] has concluded from an investigation of three cases, that the content falls within the normal range. It therefore appears advisable for us to publish the details of our results together with such extensions as have since been made.

RESULTS

For the determination of the choline-esterase, the method described by Stedman & Stedman [1935] has been used. This has been applied to defibrinated blood as well as to serum and to a suspension of washed corpuscles in physiological saline. The results have been collected in Tables I-III, which refer respectively to myasthenic patients, to patients suffering from certain other diseases and to normal subjects. The figures can be regarded as a measure of the choline-esterase content of the blood etc. employed. Actually, they represent the

Table I. *Choline-esterase in blood of myasthenics*

Date	Patient	Results		
		Blood	Serum	Corpuscles
26. ii. 35	Miss M.	0.210	0.061	0.167
4. iii. 35	Mr W. S.	0.235	0.099	0.185
12. iii. 35	Mrs M.	0.151	0.030	0.131
20. iii. 35	Mrs S.	0.188	0.089	0.140
22. iii. 35	No. 1 (from Dr Pennybacker)	0.241	0.167	0.160
30. iii. 35	No. 2 (from Dr Pennybacker)	0.190	0.089	0.136
3. v. 35	No. 3 (from Dr Pennybacker)	0.176	0.044	0.158
26. iv. 35	Mrs H.	0.181	0.038	0.156
7. vii. 35	Miss H. S.	0.206	0.100	0.168
3. iii. 36	Mr A. B.	0.201	0.093	0.134
11. vi. 36	R. W. (from Dr M. Walker)	0.139	0.070	0.090
11. vi. 36	D. C. (from Dr M. Walker)	0.145	0.102	0.096
Average		0.189	0.082	0.143

Table II. *Choline-esterase in certain pathological bloods*

Date	Disease	Results		
		Blood	Serum	Corpuscles
26. ii. 35	Neurosis	0.219	0.139	0.142
13. iii. 35	Ocular palsies	0.268	0.148	0.179
9. v. 35	Epilepsy	0.193	0.093	0.159
26. xi. 35	Polyneuritis	0.175	0.137	0.096
3. xii. 35	Progressive muscular atrophy	0.135	0.075	0.119
10. xii. 35	Progressive muscular atrophy	0.104	0.057	0.055
17. xii. 35	Muscular atrophy	0.172	0.130	0.112
30. iii. 36	Muscular dystrophy	0.168	0.075	0.145
2. iv. 36	Myotonia atrophica	0.164	0.106	0.116
20. iv. 36	Congenital myotonia	0.140	0.051	0.116
Average		0.174	0.101	0.124

Table III. *Choline esterase in normal blood*

Blood	Serum	Corpuscles
0.377	0.353	0.198
0.268	0.148	0.179
0.256	0.126	0.195
0.244	0.133	0.184
0.243	0.138	0.165
0.232	0.144	0.156
0.213	0.111	0.146
0.199	0.115	0.126
0.198	0.084	0.141
Average	0.248	0.150
		0.166

volumes of carbon dioxide liberated from bicarbonate by the acetic acid produced by the action of 0.05 ml. of the material concerned on acetylcholine for a period of 50 min. It should be emphasized that the corpuscles, after washing with saline, were brought to a volume identical with that of the blood from which they were obtained. The figures for the corpuscles therefore represent the portion of the choline-esterase activity of the whole blood for which they are responsible. The serum, on the other hand, was used in an undiluted condition. Hence the values quoted for this material will represent from 160 to 200 %, according to the corpuscular volume, of the portion of the choline-esterase activity of the whole blood which the serum contributes.

It will, we believe, at once become evident from an inspection of the individual figures in the three Tables that the results obtained with the bloods from myasthenic patients differ on the whole from those yielded both by the normals and by the non-myasthenic patients in respect of the fact that the choline-esterase activities of the sera are noticeably smaller. In order to emphasize this difference, the figures in each Table have been averaged. Thus, the average serum value for the myasthenics is 0.082 as compared with 0.101 and 0.150 for the other two groups. Nevertheless, the activities of the myasthenic corpuscles do not diverge significantly from those of the non-myasthenic groups. The myasthenic bloods are thus distinguished from the others examined in that the choline-esterase content of the serum is low as compared with that of the corpuscles, a difference which is, we believe, best illustrated by comparing the ratios of the activities of the corpuscles to those of the sera. Using average values, the figures representing these ratios are: for the myasthenics, 1.74, for the other pathological bloods, 1.23; and for the normals, 1.11. For the reasons mentioned above, these ratios do not, of course, represent the actual distribution of choline-esterase between corpuscles and serum.

In two cases of myasthenia gravis, it has been possible to estimate the choline-esterase content of the blood over an extended period of time. The actual results obtained are recorded in Tables IV and V. Added interest is attached to these results on account of the fact that, prior to the last estimation recorded, striking remissions of the disease occurred in both cases. As can be seen from

Table IV. *Variations of choline-esterase in myasthenic blood (Miss M.)*

Date	Results		
	Blood	Serum	Corpuscles
26. ii. 35	0.210	0.061	0.167
6. iii. 35	0.187	0.058	0.142
18. v. 35	0.153	0.046	0.140
25. vi. 35	0.179	0.058	0.153
25. vi. 35	0.136	0.029	(After prostigmin)
2. vii. 35	0.184	0.064	0.158
10. iii. 36	0.145	0.066	0.101

Table V. *Variations of choline-esterase in myasthenic blood (Mrs H.)*

Date	Results		
	Blood	Serum	Corpuscles
26. iv. 35	0.181	0.038	0.156
7. v. 35	0.234	0.076	0.183
7. v. 35	0.191	0.063	(After prostigmin)
14. vi. 35	0.206	0.073	0.176
1. vii. 35	0.219	0.074	0.179
8. iv. 35	0.103	0.045	0.089

the figures in the tables, the improvement in the patient's condition was, in each case, associated with a diminution in the choline-esterase content of the blood, this diminution being largely, if not wholly, due to a reduction in the content of the enzyme in the corpuscles. This result is clearly in harmony with the finding that there is, in myasthenic blood, an abnormal distribution of choline-esterase between corpuscles and serum. Incidentally, it should be pointed out that Tables IV and V give, in each case, one result which was obtained with blood

collected 1 hr. after the administration of prostigmin. The inhibitory action of the drug on the enzyme is apparent from the figures. It was not, unfortunately, possible to estimate the activity of the corpuscles under these conditions on account of the fact that during the process of washing the inhibitory effect disappeared. That the drug did, however, exercise an inhibitory action on the enzyme in the corpuscles follows from the value obtained for whole blood.

DISCUSSION

The difficulty in interpreting the above results lies in the fact that the enzyme which is effective in destroying acetylcholine is the choline-esterase which is present in the tissues. That present in the blood doubtless exists as a safeguard to deal with such acetylcholine as may escape destruction in the tissues. Nevertheless, in view of the result that the content of choline-esterase in the blood of myasthenics is undoubtedly no higher than in normals, it appears justifiable to conclude that the muscular weakness in myasthenia gravis is not due to an excessively rapid destruction of acetylcholine by the enzyme in question, but must rather be caused by a deficiency in the production of this substance in the nervous tissue or, less probably, by a defect in the mechanism of its liberation. Whether any significance can be attached to the abnormal distribution of choline-esterase between serum and corpuscles in myasthenics is not at present certain, particularly in view of the relatively few cases which it has been possible to examine. Nevertheless, it is an attractive hypothesis to consider that the concentration in the corpuscles varies with that in the tissues, or, in other words, that the factor which controls its distribution between serum and corpuscles also controls that between serum and tissues. On such an assumption one could interpret the results as indicating that the concentration of choline-esterase in the tissues of myasthenics is at approximately its normal value, and that the lowered serum value is the first result of an attempt of the body to compensate for its deficiency in acetylcholine by reducing the amount of choline-esterase available for its destruction. In the two cases in which remissions occurred, this process has apparently proceeded further and has resulted in a reduction of the choline-esterase in the tissues. If such an interpretation be justified, it would appear that the remissions are due, not to a spontaneous removal of the causes which are responsible for the muscular weakness, but to a compensatory reduction in the quantity of choline-esterase available.

SUMMARY

Estimations have been made of the choline-esterase content of blood from persons suffering from myasthenia gravis, and the values obtained have been compared with the results given by certain other pathological bloods and by normal blood. The total contents of the myasthenic and pathological bloods are, on the whole, lower than that of normal blood. The myasthenic blood is, however, distinguished from the two other groups in that the average serum content is lower, while the content of the corpuscles approximates to that of the normals. There thus appears to be a disturbance of the distribution of the enzyme between corpuscles and serum in myasthenia gravis. In two cases, striking remissions of the disease were observed to be associated with a marked diminution of the choline-esterase content of the corpuscles. A possible interpretation of these results has been advanced.

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