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Received December 17th, 1959

Biochim. Biophys. Acta, 38 (1960) 384-386

The presence of basic proteins in microsomes

The treatment of microsomes from rat liver with the detergents "Lubrol W" and sodium perfluoro-octanoate has revealed the presence of several protein fractions¹ into each of which amino acids are found to become incorporated to a different extent. Since the structure and composition of these fractions may throw light on their behaviour, in the course of the work the content of amino acids in the protein has been determined in several preparations of microsomes and their fractions. The N-terminal groups of the protein of some samples were also determined. Some amino acid analyses of the protein of whole microsomes² and of ribonucleoprotein particles³ have previously been reported. SIMKIN AND WORK⁴ gave the analyses of two fractions prepared by extraction of whole microsomes from guinea-pig liver with salt, which did not differ greatly from the whole microsome.

In the present work the protein of the microsomes of the Lubrol and PFO pellets were prepared as previously described¹. The HCl extract was obtained by treating the PFO pellet with 0.2 N HCl for 16 h at 4°. The residue was re-extracted for 6 h, and the combined extracts dialysed and freeze-dried. The total amino acid analyses of the proteins were carried out by the FDNB method of LEVY⁵ and FRAENKEL-CONRAT, HARRIS AND LEVY⁶ with the modifications described by PHILLIPS AND JOHNS⁷. The N-terminal groups were determined according to the technique of PHILLIPS⁸.

Table I gives the results of complete amino acid analyses of samples from three different preparations. The proportions of various N-terminal groups of some of the samples can be seen in Table II. The results show that the total protein of the microsomes is not predominantly basic since the sum of the acidic amino acids is greater than that of the basic. However, there is some tendency for the content of basic amino acids to increase in the Lubrol and PFO pellets. The HCl extract of the PFO pellet shows a marked increase of the basic residues indicating that there are among the microsomal proteins some which are markedly basic. The total percentage of basic amino acids in this fraction is 27.8, which is higher than that normally found in histones.

Abbreviations: PFO, perfluoro-octate; FDNB, fluorodinitrobenzene.

As regards end-groups, it is remarkable to find that proline and alanine combined form almost 40 % (or even more) of the N-terminal groups both of the whole microsomes and of the Lubrol and PFO pellets. This may be compared with the results for histones of calf thymus in which these two N-terminal groups comprise at least 80 % of the total amount⁷. The main difference observed in the microsomes and their fractions is the high proportion of N-terminal glycine groups (25–33 %). As the latter may occur in a distinct protein, it is likely that the microsomes contain proteins similar

TABLE I
AMINO ACID COMPOSITION OF THE PROTEIN OF WHOLE MICROSOMES AND
MICROSOMAL FRACTIONS
(moles % of amino acids determined)

<i>Amino acid</i>	<i>Whole microsomes I</i>	<i>Whole microsomes II</i>	<i>Lubrol pellet II</i>	<i>PFO pellet II</i>	<i>PFO pellet III</i>	<i>HCl extract of PFO pellet III</i>
Aspartic acid	8.8	10.1	9.1	8.0	8.3	7.5
Glutamic acid	12.7	11.5	10.7	11.2	10.6	10.4
Alanine	7.6	7.4	8.1	7.9	7.6	8.5
Valine	8.4	7.5	21.7	8.3	8.5	6.8
Leucine/isoleucine	13.9	15.7		14.9	13.7	11.7
Serine	6.3	6.1	6.6	6.3	5.2	5.3
Threonine	5.2	5.5	5.4	5.4	4.9	4.6
Glycine	7.3	6.8	7.5	7.4	9.4	7.5
Tyrosine	1.4	1.4	2.2	2.1	2.4	2.1
Phenylalanine	5.0	5.8	5.1	5.3	5.2	3.0
Methionine	1.0	—	0.7	—	—	—
Proline	5.9	5.9	5.6	5.4	4.7	4.8
Histidine	2.2	2.8	2.6	2.0	2.8	2.7
Lysine	6.6	6.8	8.0	8.4	8.1	13.6
Arginine	7.6	6.8	6.7	7.3	8.5	11.5

TABLE II
N-TERMINAL GROUPS OF THE PROTEIN OF WHOLE MICROSOMES AND MICROSOMAL FRACTIONS
(proportions of N-terminal groups expressed as percentages of the total end-groups found)

<i>Amino acid</i>	<i>Whole microsomes II</i>	<i>Lubrol pellet II</i>	<i>PFO pellet II</i>	<i>PFO pellet III</i>
Aspartic acid	10.2	3.1	3.2	3.0
Glutamic acid		2.7	2.6	2.6
Alanine	8.1	10.3	13.5	15.4
Valine	2.8	3.4	0.3	3.0
Leucine/isoleucine	2.8	2.0	2.4	2.1
Serine	10.0	8.0	5.0	8.1
Threonine	6.2	5.2	4.5	3.0
Glycine	25.1	31.1	33.1	28.7
Tyrosine	—	—	2.1	—
Phenylalanine	1.2	0.9	2.6	—
Proline	31.0	29.0	25.6	28.7
Histidine	—	—	0.5	—
Lysine	2.7	4.2	4.2	5.6
Arginine	—	—	—	—

in composition to the histones found in the nucleus. Further fractionations are being attempted.

This investigation has been supported by grants to the Chester Beatty Research Institute (Institute of Cancer Research: Royal Cancer Hospital) from the Medical Research Council, the British Empire Cancer Campaign, the Jane Coffin Childs Memorial Fund for Medical Research, the Anna Fuller Fund, and the National Cancer Institute of the National Institutes of Health, U.S. Public Health Service.

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Received January 7th, 1960

Biochim. Biophys. Acta, 38 (1960) 386-388

Announcement

5th Meeting of the International Conference on Biochemical Problems of Lipids

The 5th Meeting of the International Conference on Biochemical Problems of Lipids will take place in Marseilles (France) on July 21-23, 1960. Subject: "The enzymes of lipid metabolism". Sections: (1) Techniques, (2) Hydrolysis, (3) Oxidative degradation, (4) Biosynthesis.

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