THE STRUCTURE OF LACTOSOMATOTROPIC HORMONE

IV. PEPTIDES OF TRYPTIC HYDROLYSIS OF THE FRAGMENTS D-1, GD-2, G-1, AND E-1 OF LSTH

N. D. Gafurova and F. Yu. Ryshka

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The lactosomatotropic hormone (LSTH) has been subjected to cyanogen bromide cleavage [1]. A study of the resulting fragments showed a considerable similarity of the structures of LSTH and ovine lactogenic hormone (O-LTH) [2]. The present paper gives the results of the separation and characterization of the products of enzymatic hydrolysis of the cyanogen bromide fragments G-1, D-1, GD-2, and E-1 of LSTH, and also a comparison of the results obtained with the known structure of O-LTH.

Fragments G-1, D-1, and GD-2, containing basic amino acids, were hydrolyzed by trypsin. The peptides were separated by partition chromatography in a column of cellulose in various solvent systems. Fragment E-1 was hydrolyzed by chymotrypsin and the resulting peptides were separated by preparative paper electrophoresis. The homogeneity of all the peptides isolated was checked by chromatoelectrophoresis in a thin layer of cellulose. The amino-acid compositions of the peptides investigated were determined

TABLE 1. Amino-acid Compositions and N-Terminal Amino Acids of the Peptides from the Enzymatic Hydrolysis of the Fragments G-1, D-1, GD-2, and E-1 of LSTH

Amino acid	Fragment D-1			Fragment GD-2			Fragment E-1		Fragment G-1				
	DIT-1	DIT-2	DIT-3	GDT-1	GDT-2	GDT-3	EIX-1	EIX-2	GIT-1	GIT-2	GIT-3	GIT-4	GIT-5
Lys											1,0(1)	1,1(1)	1,0(1)
His	1,1(1)						1,2(1)	1,1(1)					
Arg	1,1(1)	1,0(1)		1,1(1)	1,0(1)					0,8(1)	1,0(1)	1,0(1)	
Asx	2,0(2)			2,1(1)	2,0(1)			1,0(1)		1,1(1)	1,0(1)	1,0(1)	
Thr	0,8(1)			0,8(1)									
Ser	0,9(1)	0,9(1)		0,9(1)			1,0(1)	1,9(2)		1,0(1)	1,0(1)		
Glx	1,0(1)			1,0(1)				0,9(1)	1,0(1)			4,0(4)	
Pro	1,2(1)			3,2(3)						0,9(1)	0,7(1)		
Gly		1,0(1)	1,0(1)	2,0(2)					1,0(1)	1,3(1)	1,3(1)		
Ala						1,0(1)				1,8(2)	2,0(2)	1,1(1)	
Cys*				(2)									
Val	1,8,2)			1,7(2)		0,9(1)	0,8(1)						
Met†			(1)			(1)		(1)	(1)				
Ile		0,7(1)						0,7(1)		0,9(1)	1,0(1)	2,0(2)	
Leu	2,0(2)	3,0(3)		1,0(1)	0,9(1)			0,9(1)	1,8(2)	1,2(1)	0,9(1)		
Tyr	0,8(1)						0,7(1)						
Phe					0,9(1)								
Trp	(1)												
No. of residues	14	7	2	16	5	3	4	8	5	9	10	10	1
N-terminal amino acids	Ser	Ser	Gly	Thr	Asx	Ala	Val	Ile	Leu	Gly	Lys	Ala	

*To determine the cysteine, before hydrolysis the peptides were oxidized with performic acid.
†The methionine was determined from the content of homoserine lactone.

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on an automatic amino-acid analyzer. The only tryptophan fragment of D-1 was found in the peptide DT-1 by the qualitative Ehrlich reaction. The N-terminal amino acids were determined by the reaction with dansyl chloride (DNS-Cl) followed by the chromatography of the DNS derivatives in a thin layer of silica gel or on polyamide plates [3, 4]. The stepwise degradation of peptide GT-2 was performed by Edman's method [5] (Table 1).

The peptides isolated from the fragments D-1, GD-2, and E-1 coincided completely with sections of the structure of O-LTH 1-36 and 82-104. In the G-1 fragment, in the third position from the N-end, as compared with section 105-129 of the structure valine has been replaced by alanine. The other peptides, the amino-acid sequences of which were not studied, can differ from the known structure of O-LTH only by a rearrangement of the amino-acid residues.

In a study of the structure of bovine LTH (B-LTH), from a tryptic hydrolyzate we isolated a peptide corresponding to the sequence 106-114 of O-LTH, but with the replacement of the valine in position 107 by alanine [6, 7]. Peptide chains 115-124 and 125-129 of the two hormones had identical amino-acid compositions.

Thus, fragment G-1 is probably identical in structure with section 105-129 of the B-LTH molecule. The substitution observed is apparently species-specific and characteristic of the two hypophyseal hormones of cattle, LSTH with LTH, which have similar structures.

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