DETERMINATION OF THE C-TERMINAL AMINO ACID RESIDUES OF THE PROTEINS OF THE HORMONES OF THE HYPOPHYSIS

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TABLE 1. C-Terminal Amino Acids of Some Hormones

Hormone	C-Terminal amino acids	Other amino acids split off by carb- oxypeptidase A
STH	Phenylalanine	Not found
LST	Glycine	Ser, Leu, Ala, Val
LTH	Not found	Not found
phe-LH	Serine	Tyr, Leu, Gly, Ala
threo-LH	Serine	Asp, Tyr, Leu, Ile
FSH	Leucine	Ala, Ser, Tyr, Phe
TSH	Alanine	Leu, Ser, Tyr, Phe

We have determined the C-terminal amino acid residues of a number of protein hormones from bovine hypophyses: somatotropic hormone (STH); lactosomatotropic hormone (LSTH); luteotropic hormone (LTH); thyroid-stimulating hormone (TSH); phe-LH - the phenylalanine hormone; threo-LH - the threonine hormone; and follicle-stimulating hormone (FSH).

The preparations of LSTH, LTH, phe-LH, FSH, and TSH were obtained by extracting the hypophyses with acidified acetone [1] and the preparations of threo-LH and STH by aqueous salt extraction [2]. Further purification was carried out by ion-exchange chromatography and gel filtration.

The C-terminal amino acid residues were determined by a modification of a well-known method [3]. The hormone concerned (0.5-1 mmole) in a 0.02 M solution of ammonium bicarbonate containing dodecyl sulfate was incubated at pH 7.8-8.0 with carboxypeptidase A of the Reanal firm (ratio of enzyme to substrate 1:30) at 37°C. After predetermined intervals of time (10 min-24 h), aliquots were taken, the reaction in the sample was stopped by bringing the pH to 1-2, and the precipitate that had deposited was separated off by centrifuging. The amino acid composition of the supernatant fraction was determined on a Beckman 120C amino acid analyzer.

It can be seen from Table 1 that STH, LSTH, and LTH, which have similar biological properties, differ in their C-terminal amino acid residues. The phe-LH and threo-LH have the same C-terminal residues (serine) but the subsequent amino acid sequences in them are apparently different.

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