THE AMINO ACID SEQUENCE OF EQUINE GROWTH HORMONE

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1. Introduction

Chemical and physicochemical data supporting the existence of a close analogy between equine and other mammalian growth hormones has recently been published [1]. The structural reasons for these findings have started to emerge through the sequence study of a pentadecapeptide from the C-terminus of equine growth hormone [2] and of a 62-amino acids fragment which included the two disulphide bridges in the molecule [3]. In both instances a high degree of homology was found with bovine growth hormone.

In this paper we report the complete primary structure of equine growth hormone. The amino acid sequence of a number of peptides is partially based on homology with bovine growth hormone.

2. Materials and methods

2.1. *Equine growth hormone* was obtained as indicated in [1]. Other materials were as in [3].

2.2. *Tryptic peptides* of native, oxidized and aminoethylated equine growth hormone were obtained as previously described [3].

2.3. *Isolation of a tryptophan-containing peptide* was performed as indicated by Fernández et al. [4], but after the electrophoresis at pH 6.5 the peptide was purified by chromatography on paper developed with butan-1-ol–formic acid–water (45:45:10, by vol).

2.4. *Chymotryptic peptides* were obtained and fractionated by ion-exchange chromatography, as described in [5].

2.5. *Cystine-containing peptic peptides* were obtained as indicated in [3]. Other peptic peptides were purified by peptide mapping [3].

2.6. *Cyanogen bromide attack* was performed according to the procedure described for bovine [6] and ovine [7] growth hormones. The lyophilized reaction product was dissolved in 50% acetic acid and transferred to a Sephadex G-75 column (2.5 cm × 90 cm) equilibrated with 50% acetic acid. The elution was carried out with the same solvent at a rate of 20 ml/hr; fractions of 3 ml were collected and aliquots from every third tube were analyzed after hydrolysis in a Technicon TSM-1 autoanalyzer.

3. Results and discussion

All the information collected about the primary structure of equine growth hormone is organized in fig. 1 to give a unique sequence of amino acids. The previously published [3] structure of the disulphide bridges region is included. The single tryptophan residue is in position 85. The four methionine residues are in positions 4, 72, 123 and 178.

The complete sequences of bovine, ovine and human growth hormones are also included in the same figure.

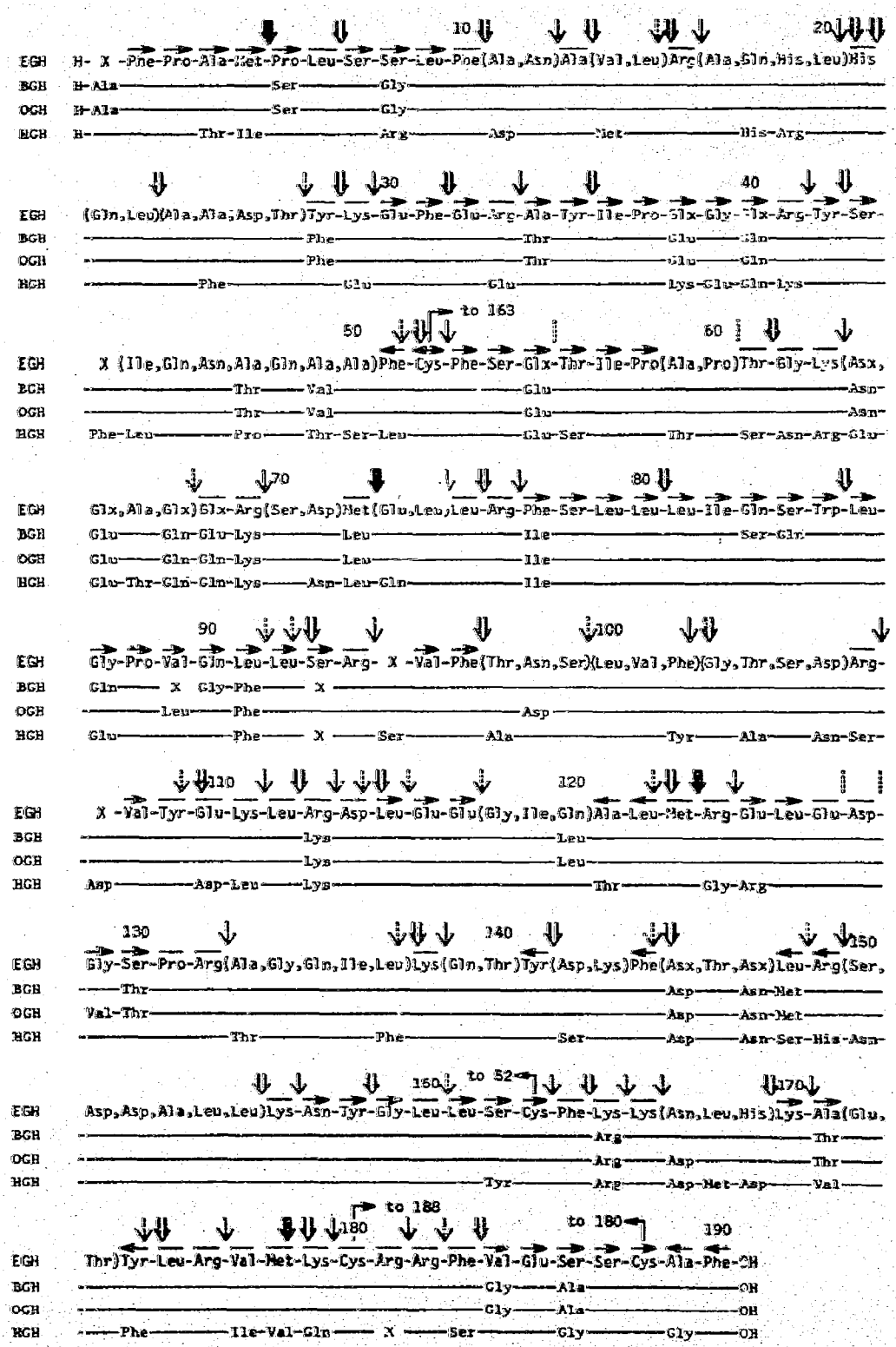

Fig. 1. Complete amino acid sequence of equine growth hormone. Primary structures of bovine, ovine and human growth hormones are included for comparison. The sequences are aligned so as to present maximum homology. Horizontal lines indicate identity with equine growth hormone. Parentheses are used to show that the identity is based on amino acid composition only. Deletions are indicated by an X. The state of amidation of glutamyl and aspartyl residues could be decided in some cases, from the electrophoretic mobility of the corresponding peptides. ↓, Points of tryptic attack; ⇓, points of chymotryptic attack; ⇣, points of pepsin attack; ↓, points of attack of cyanogen bromide; ¦ ¦, partial acid hydrolysis. →, Steps of Edman degradation; ←, residues released by carboxypeptidase A or B; —, amino acids whose sequence was established by overlapping of peptides, specific cleavage or by difference. Data are taken as follows: Equine growth hormone, the present paper; bovine growth hormone: [5]; ovine growth hormone: [4, 8]; human growth hormone: [9, 10].

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