

**Cloning and DNA sequence analysis of the cDNA for the precursor of ovine follicle stimulating hormone  $\beta$ -subunit**

Peter S.Mountford, Paul A.Bello, Malcolm R.Brandon and Timothy E.Adams

Department of Veterinary Preclinical Sciences, The University of Melbourne, Parkville, Victoria 3052, Australia  
 Submitted July 3, 1989  
 EMBL accession no. X15493

Clones coding for the  $\beta$ -subunit of ovine follicle stimulating hormone (FSH) were selected from an ovine lamb pituitary  $\lambda$ -gt10 cDNA library using two synthetic oligonucleotide probes. Both probes were synthesised on the basis of published amino acid sequence for the FSH  $\beta$ -subunit [1]. The complete DNA sequence for the ovine  $\beta$ -FSH clone 6.14, determined by dideoxy sequence analysis [2] and exonuclease III digestion [3], is presented in Figure 1.

The clone is 1589 base pairs in length and includes the entire coding region for a 129 amino acid FSH  $\beta$ -subunit prehormone. The signal peptide is 19 amino acids in length. When the predicted amino acid sequence is compared with that previously established by amino acid sequencing [1], four conservative substitutions and a single C-terminal amino acid exclusion were noted. The predicted amino acid sequence was confirmed by DNA sequencing of two independent cDNA clones. Comparison of the ovine nucleotide coding region sequence with the corresponding bovine sequence [4] gives an overall homology of 95%. Comparison of the entire ovine  $\beta$ -FSH cDNA clone 6.14, which includes a 149 bp 3' non-coding region inclusion, gives an overall homology of 85%. Subsequent sequence analysis of an additional  $\beta$ -FSH cDNA clone has revealed an additional 3' extension of approximately 300 bp not seen in clone 6.14. The extension appears closely homologous to the equivalent published bovine nucleotide sequence.

```

                                     -19      H K S V Q F C F L F C C W R A I C C R 1 S C E
ACAAGGACGCTGTCTACGGAAGGCTCAGCAGTCCACAGTTACCAAGTGCCAGGATGAAGTCCCTCCAGTCTGCTTCTCTTTCTGTGCTGGAGAGCAATCTGCTGCAGAAAGTGGCA 120
L T N I T I T V E K E E C S F C I S I N T T W C A G Y C Y T R D L V Y K D P A R
GCTGACCAACATCACCATCACGGTGGAGAAAGAGGAATGTAGCTTCTGCATAAGCATCAACACCAGTGGTGTGCGAGGCTATTGCTACACCCGGGACTTGGTGTACAAGGACCCAGCGAG 240
P N I Q K A I C T F K E L V Y E T V K V P G C A H H A D S L Y T Y P V A T E C H C
GCCCAACATCAGAAAGCATGTACCTTCAAGGAGCTGGTGTACGAGACGGTGAAGTGCCTGGCTGTGCCACCATGCAGACTCCCTGTACACGTACCCAGTAGCCACTGAATGTCACTG 360
G K C D R I D S T D C T V R G L G P S Y C S F S D I R I E I *
CGGCAAGTGTGACCCGGACAGCACTGACTGCACCGTGGCAGGGCTGGGGCCAGCTACTGCTCCTTCAGTGACATCAGAGAATAAAGAGCAGCAGCTGCTTTGAGCTGCCTACCCCTAAA 480
GGACAAAACATCCAAAGTGTCTGTGTACATCGCCCTAGGCTGCAGACCACCAAGAACCTACTGATCTCTGCTCTCTGAGAGGGGGAGCTCCAGGAATGGAGAGGGCTGGGGC 600
CTGGCACCACACAGACCTGTATTCTACATCTGGTTCATGAGTCTTACTCACTCTCACTTAACTTACAGACAGAGGGTGTCTTTCCATTTAATACTTAGAATCTCTCAGGCAATC 720
CCTTCTCTTAAAGCTAGGATACGGTCCACAGGGGAAGGAATGAGCTAAAAGTAGGTGAAAGTCTTAAAGTGCAGCATCTCTCTGCGAGACTCAAGAGCCCTCAAGAGAAGGCCAAGCT 840
CTTCTCATGATTGTAGCCTCAATGCCTAACACTCCACATAGAAATTTAGTAAGAACTTAAATAAGGCTTCTTAAAGAAAGTAAAGTACAGAAAGTTAGGGATAGGAGAACACTGAGAGA 960
GGATGTTAGAGGCTATCTGATAGGGCTCGCCTCCAGGGAAGGGGGGCATCAGCTTCACTTCTGCTCTGCTCTCTCTCTGCGCAGATGGAGGATGGAGACAGGATAAACTCTCTGAAT 1080
CATTTCAGGAGGTGGCAGTTACCAAGCTCAGGATTTCAAGCTATTACTGCCAGTCTTTAGTTAAGGGCAAAGCAAGAATATAATACGGGTTTGTGGAAATTAGCTGCATCTATT 1200
CACCTTTTCACTTTTCATGCATTTGGAGAAGGAAATGGCAACCCTCCAGTGTCTTTCCTGGAGAAATCCAGGGACGGGGAGCTTGGTGGGCTGCCATCTATGAGGTTGCAGAGGGTCCG 1320
ACACGACTGAAGTGACTTAGCAGCAGCATCCATCATTTTAAATAAATGGAACAAATGCTGTGAGGCTCCTGCAAAAACCTTTCCGGGAAGTACATAGATCTATATCACCCCTGCCCTTGAAT 1440
GTAGGCTCTACTCCTTTGGAAAAGGTTGGATTTGAGAGCAGGTTTGAAGCAATTTTGAACACTTAATAAGTACATTTATCTTATCTCAAAATACATTTATGTAAAGAAAATGCTCTTT 1560
AGAAAGCAATTAGCCATACAAAAAAAAAA 1589
    
```

**Figure 1.** Nucleotide sequence and predicted amino acid sequence of the ovine FSH  $\beta$ -subunit precursor hormone cDNA clone 6.14. The position of the C-terminal amino acid exclusion is immediately prior to the termination codon (indicated by the asterisk). The 149 bp inclusion is underlined. The amino acids differing from the published peptide sequence [1] are indicated by †.

**References**

- [1] Sairam, M.R., Seidah, N.G. and Chretien, M. (1981) *Biochem. J.* **197**: 541-552.
- [2] Sanger, F., Nicklen, S. and Coulson, A. R. (1977) *Proc. Natl. Acad. Sci. USA.* **74**: 5463-5467.
- [3] Henikoff, S. (1984) *Gene* **28**: 351-359.
- [4] Maurer, R.A. and Beck, A. (1986) *DNA* **5**: 363-369.