

The nucleotide and deduced amino acid sequences of insulin-like growth factor II cDNAs from adult bovine and fetal sheep liver

W.M.Brown, K.M.Dziegielewska, R.C.Foreman and N.R.Saunders

Department of Physiology and Pharmacology, The University of Southampton, Bassett Crescent East, Southampton SO9 3TU, UK

Submitted June 25, 1990

EMBL accession nos X53553, X53554

Using immunological screening a partial insulin-like growth factor II cDNA clone was isolated from an adult bovine liver library in λ gt11 (Clontech). The clone (EMBL accession number X53553) encodes most of the B chain, the whole of the C, D and E peptides and the A chain of prepro IGF-II (for nomenclature see 6).

The deduced (partial) amino acid sequence of mature IGF-II (i.e. without signal peptide or the E domain) is in complete agreement with the protein sequence reported in (2) and differs only by one substitution from that reported in (3) – we observe *ile* at position 35, not *ser*.

The bovine clone was used to screen a fetal sheep liver (pooled 40–60 day tissue) cDNA library in λ gt10. A full length clone was isolated encoding the whole of prepro IGF-II. The sequence of both clones was determined by the dideoxynucleotide method (5).

The sheep clone (EMBL accession number X53554) is significantly longer than that reported in (4), the 3'-untranslated region (UTR) being some 70 bases longer and 5'-UTR over 200 bases longer and of a different sequence. The coding sequence is, however, identical.

The IGF-II gene has been extensively studied in recent years, and it is now clear that multiple transcripts arise from a single-copy gene, by use of three different promoters (1, 4, 6 and references therein). In the rat, there is a splice site six bases 5'- to the initiation codon, and alternative transcripts arise as a result

of 3 different exons being spliced at this point to the first coding exon E4 (1). Our sequence deviates from that in (4) seven bases 5' to the initiation codon. It, thus, seems likely that a similar splicing mechanism operates in sheep. By comparison with the rat sequences reported in (1), it appears that the 5'-UTR reported here corresponds to rat exon E2.

Both sequences are presented. Numbers to the right refer to nucleotides, to the left to the amino acid sequence of the mature IGF-II, the putative signal peptide having negative numbers. The extent of mature IGF-II is indicated by underlining. The E domain is enclosed by square brackets. The termination codon is indicated by an asterisk (*).

ACKNOWLEDGEMENTS

This work was funded by grants from The Wellcome Trust, Action Research for the Crippled Child and The Wessex Medical Trust. We thank Dr. I.G.Giles for his invaluable advice.

REFERENCES

1. Ueno, T. *et al.* (1988) *Biochim. Biophys. Acta* **950**, 411–419.
2. Francis, G.L. *et al.* (1988) *Biochem. J.* **251**, 95–103.
3. Honégger, A. and Humbel, R.E. (1986) *J. Biol. Chem.* **261**, 569–575.
4. O'Mahoney, J.V. and Adams, T.F. (1989) *Nucl. Acids Res.* **17**, 5392.
5. Sanger, F. *et al.* (1977) *Proc. Natl. Acad. Sci. USA* **74**, 5463–5467.
6. Dull, T.J. *et al.* (1984) *Nature (London)* **310**, 777–781.

Bovine clone, Accession No. X53553.

```

6  E T L C G G E L V D T L Q F V C G D R G F Y F S R P S S R I N R R S R G
   CGAGACTCTGTGGGGGGGAGCTGGTGGACACCTCCAGTTTGTCTGTGGGACCGGGCTTCTACTTCAGCGACCATCCAGCGCATAAACCGAGCGACGCGTGGG 109
42  I V E E C C F R S C D L A L L E T Y C A T P A K S E [ R D V S A S T T V L
   ATCGTGGAAAGTGTTCCTCCGAACTGGGACCTGGCCCTGGAGACTACTGTGCCACCCCGCAAGTCCGAG AGGGATGTCTGCCTCTACGACCGTGCCTTC 218
   P D D V T A Y P V G K F F Q Y D I W K O S T Q R L R R G L P A F L R A R
   CGGACGAGTCCACCGATACCCCGTGGCAAGTTCTTCCAATATGACATCTGGAAAGCAGTCCACCCAGCGCTCGCGAGGGGCTCCCGCCCTTCCCTGGGAGACGCCG 327
   G R T L A K E L E A L R E A K S H R P L I A L P T O D P A I H G G A S S
   GGTCCACCGCTCCCAAGGAGCTGGAGGCCCTCAGAGAGGCCAAGAGTCCCGCTGATCGCCCTGCCACCCAGGACCTGCCATCCAGGGGGGCCCTCTTC 436
   K A S S D *
   AAGGCATCCAGCGATTAG AAGTGAAGCAAGTGTGTAATCTGCCAAGTGGCACCATCTACCTCGGGCGACCTCTCGACCGGACCGCCCACTAGGTCTCTCTCTG 545
   AAA TCCCTGTACCGTCC TGTCTGGGGCTCCCTTGGCCCGGCCCTGTGTCGCCCAACC TCCCGAGTCAAGGCGAATCCCGCTCGGCCCTCCATCTGCTGAGGGGATC 654
   AGAACAATCTCTAAAATGTACAAAACCAATTTGGCTTTAAATATCCCCCAAAATATACCCCAAAATACCCCAAAATACACCAACCAAAATGCAATCATGAAC 763
   CCTCAATCAGCCCTTGAACGAATTTGGCTTTTAGCAACACCGAAAAGCAACTAGCTTTCCAAAATCTCTAAAAC 845

```

Ovine clone, Accession No. X53554.

```

GCCCGCCCGCCCGCCGACAGCCCGTCCCTTAAAGAAATCAGAGAAAATTTCCCGCCCGCCCAAAAACAGCCCAATCCCGCGTCTCGCCCGTCCGACATTCGGCC 109
   CCGCGAGCTCGGTTCAGAGCGCCCTCGCAGAGGAGTGGCCCGCAGGACCTTCCGCGCTGTTCGGTTTGCATACCGACAGGAGAGTGGCGGCTCGGTCCGCGCTT 218
-24  H G I T A G K S M L A L L A F L A F A S C C Y A A Y R P S E T L C
   CCAGACATCAATGGGATCAGCAGCAAGTCGATCTGGCCCTTCTGTCTTGGCCCTCGCTGCTATGCTTCTACCGCCCGCCAGCAGACTGTGTC 327
10  G G E L V D T L Q F V C G D R G F Y F S R P S S R I N R R S R G I V E E
   GCGGGGAGTGGTGGACACCTCCAGTTTGTCTGTGGGACCGGGCTTCTACTTCAGCGACCATCCAGCGCATAAACCGAGCGACCGCTGGCATCTGGAAAGAT 436
46  C C F R S C D L A L L E T Y C A A P A K S E [ R D V S A S T T V L P D D F T
   GTTCTTCCGAACTGGCAGCTGGCCCTCTGGAGACTTACTGTGCCCGCCCGCAAGTCCGAG AGGGATGTCTGCCTCTACGACCGTCTCCGACGAGCTTCAC 545
   A Y P V G K F F Q S D T W K O S T Q R L R R G L P A F L R A R R G R T L
   AGCATACCCCGTGGGCAAGTTCTTCCAATCTGACACCTGGAAAGCAGTCCACCCAGCGCTCGCGAGGGGCTCCCGCCCTTCCCTGGGAGACCGCGGGCTCCGACGCTC 654
   A K E L E A L R E A K S H R P L I A L P T O D P A T H G G A S S E A S S
   GCCAAGGAGCTGGAGGCCCTCAGAGAGGCCAAGATCCCGCTGATCGCCCTGCCCTACCCAGGACCTGCCACCCAGGGGGGCCCTTCTCCGAAAGCATCCAGGC 763
   D *
   ATTAG AAGTGAAGCAAGTGTGTAATCTGCCAAGTGGCACCATCTACCTCGGGCGACCTCTCGACCGGAC TGGCCCAATGGTCTCTCTGAAATCCCTGTACC 872
   GTCTGTCTGGGGGCTCCCTTACCCAGCTCTCTGCCCAACCTCCCAAGTCAAGGAGCTTCTCGCGCCCTCCATCTCGCCGAGGGATCAGAACAATATCTCA 981
   AAAATGTACAAAACCAATTTGGCTTTAAATATCCCCCAAAATATACCCCAAAATATACCAACCAAAATGCAATCATGAACCTCAATCAGCCCT 1090
   CTGTAAGAAATTTGGCTTTTAGCAACACCGAAAAGCAACTAGCTTTCCAAAATCTCTAAAACAAAAA 1167

```