

# Expression and secretion of the human placental growth hormone in *Escherichia coli*

A.Igout, M.L.Scippo, F.Frankenne and G.Hennen

Laboratoire d'Endocrinologie, Institut de Pathologie, CHU B23, B-4000 Liege, Belgium  
Submitted April 20, 1989

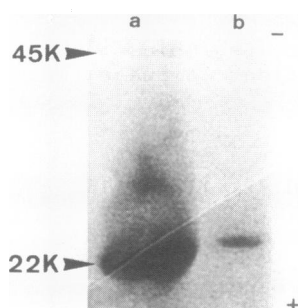
The GH-V gene is expressed during human pregnancy in the placenta (1). It codes for human placental growth hormone (HPGH)(1), which is secreted in maternal blood where it replaces pituitary GH in late pregnancy (2). We have recently cloned and sequenced the GH-V cDNA (3) and we report here on its expression in *E.coli*.

After modification using synthetic oligonucleotides, this cDNA was inserted in the pINIII-ompA3 plasmid (4) in order to have the GH-V protein secreted in the periplasmic space. *E.coli* D1210 (5) was transformed with the constructed plasmid pIPGH1. The GH-V cDNA expression was induced for 3 hr by isopropyl- $\beta$ -D-thiogalactopyranoside (IPTG) and the periplasm content was separated from the cytoplasm fraction after osmotic shock (6). Two anti-GH MAbs (5B4 and K24)(2) and an anti-GH antiserum were used to characterise the expression product.

The 5B4 MAb reacts with the N-terminal part of GH and recognizes pituitary and placental GH, the K24 reacts with an internally located epitope and recognizes pituitary but not placental GH (2). The antiserum reacts with both

variants. From the RIA data, it appears that the periplasm contains a factor reacting with the 5B4 MAb and with the anti-GH antiserum but not with the K24 MAb. The antigen was revealed by western blotting (figure 1) as a 22K protein, the expected size of the GH-V protein.

The immunoreactivity pattern as well as the molecular size of the secreted GH protein demonstrates the expression and secretion of recombinant GH-V protein and its similarity to the native hormone placental GH.



**Fig.1:** Western blot of the periplasmic fraction (track b) compared to pituitary GH (track a). The anti-GH 5B4 MAb was used as specific antibody.

## References

- 1 Scippo, M.L., Frankenne, F., Van Baeumen, A. Igout and G. Hennen. Arch. Int. Physiol. Biochim., 1989, 97, B59.
- 2 Frankenne, F., Closset, J., Gomez, F., Scippo, M.L., Smal, J. and Hennen, G. J. Clin. Endocrinol. & Metab., 1988, 66: 1171-1180.
- 3 Igout, A., Scippo, M.L., Frankenne, F. and G. Hennen. Arch. Int. Physiol. Biochim., 1988, 96: 63-67.
- 4 Ghayeb, J., Kimura, H., Takahara, M., Halung, H., Masui, Y. and Inouye, M. EMBO J., 1984, 3: 2437-2442.
- 5 Salder, J.R., Tecklenburg, M. and Betz, J.L. Gene, 1980, 8:279-300.
- 6 Keshland, D. and Batstein, D. Cell, 1980, 20, 749-760.