

## **Recombinant cytokine production at Moredun Research Institute**

Recombinant ovine and bovine cytokines have been expressed using a mammalian expression system. Cytokines are secreted into the cell culture medium of Chinese hamster ovary (CHO) cells transfected with a cytokine gene or cDNA, encoding a gene, using the pEE14 Glutamine Synthase (GS) Expression System™ (Lonza). The GS Expression System™ has been selected with a view to producing biologically-active cytokines in sterile tissue culture without addition of antibiotics.

Following selection, cloned lines of transfected CHO cells are established and maintained in Glasgow's Modified Eagle's Medium (GMEM) supplemented with sodium pyruvate, non-essential amino acids and pestivirus-free USDA-approved dialysed fetal calf serum (FBS).

For cytokine production, CHO cells are cultured in FBS-free GMEM and without selection marker (methionine sulphoxamine). The cell-free CHO supernates are checked for *Mycoplasma* spp. and endotoxin contamination. Supernate from untransfected CHO cells is recommended as a negative control for cytokine activity in bioassays and for verifying specificity of ELISAs.

Estimates of protein concentrations and biological activities are provided where known. Cytokine concentrations may vary on a batch-to-batch basis but can be adjusted for experimental use. These reagents are produced by the Moredun Immunology Theme in conjunction with the BBSRC/RERAD Immunological Toolbox and are for *in vitro* use only. Further information on cytokine activity and their availability (on a collaborative basis) can be obtained from Sean Wattedgera and Gary Entrican at [cytokines@moredun.ac.uk](mailto:cytokines@moredun.ac.uk).

### **References:**

Bebbington, C.R. (1995). Use of vectors based on gene amplification for the expression of cloned genes in mammalian cells. *DNA Cloning 4, Mammalian Systems*. Oxford University Press, UK.

Cockett, M.I., Bebbington, C.R. & Yarranton, G.T. (1990). High level expression of tissue inhibitor of metalloproteinases in Chinese hamster ovary cells using glutamine synthetase gene amplification. *Biotechnology* **8**, 662-667.