



Protein Production Service from Genaxxon BioScience

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The Company

Genaxxon is based in the south-west of Germany with a subsidiary in Munich. We are specialised on the production of recombinant proteins (DNA-polymerases – extracellular matrix proteins). Our customer base is from Pharma, Biotech and academic institutions. Being entirely dedicated to production of proteins and distribution of products for research customers, we do not perform any internal drug discovery activities.

As Genaxxon is part of a net-work of different companies with expertise in protein production, we can also offer a high throughput crystallisation screen service that is performed in nanolitres scales, with only low amounts of protein needed. Results for your protein of interest will be delivered at a fixed price with a typical turn around time of two weeks. Customer receives a crystallisation summary that lists all conditions used. As a separate module images of all wells are available. If wished crystals obtained from the screen(s) can be delivered.

Recombinant Protein Production Service

The protein of interest can be cloned and expressed in E.coli or insect cell systems. If fully glycosylated proteins are needed we will transfect human cell lines with cloned DNA and grow the human cells under reproducible serum free conditions, allowing to express molecules which are very close to those existing naturally. Production in serum free environment applies to the standards for hygiene, abstinence of viruses, function and purity of the product. In all cases we will apply state of the art technologies offering basic researcher as well as tissue engineers products of constant high quality.

Recombinant production in a human cell line enables to produce products with a processing and glycosilation pattern close to that in vivo. A considerate purification conserves the tertiary structure and consequently the biological activity.

Depending on the needs of our customer our recombinant proteins from E.coli or insect cells are tested in house for crystallisation and can be certified as crystal grade. Customer receives the protein and a detailed information sheet about protein features and crystallisation conditions.

Usual batch sizes are 5 mg and 10 mg. Other sizes are available on request.

Standard prices are in the range of 7500,00 Euro per 5 mg and 12500,00 Euro per 10 mg of crystal grade proteins.



Crystallisation Service - Prices

Technology Platform

Three dimensional structures of target compound complexes are unique sources of information during the rational drug discovery process. Straightforward availability of structural data at an early stage of the drug development process significantly enhances your productivity and success rates during hit selection, lead generation, and lead optimisation.

Available Screens

Screens are generally performed in 96 well plates where each well holds a maximum of three protein drops.

Initial Screen (4 possible set-ups are available)

- 1 Protein at one concentration (100,00 Euro per plate)
- 1 Protein at 3 different concentrations (200,00 Euro per plate)
- 1 protein at 3 different drop compositions (200,00 Euro per plate)
- 3 different proteins (200,00 Euro per plate)

Additive and Detergent Screen

Crystallisation enhancing components are added to the screen
(200,00 Euro per plate)

Compound Screen

A series of compounds is screened for binders and non-binders
(100,00 Euro per plate)

Ionic Liquid Screen

Ionic Liquids are added to stabilise and enhance crystal growth
(200,00 Euro per plate)

Co-Crystallisation of Ligands and Compounds

Classically applied to find optimal conditions for one ligand/compound
(200,00 Euro per plate)

Image Series

Images of all wells to check crystals growth
(25,00 Euro per plate)



Recombinant Protein Production in Human HEK293 Cells

Are you facing problems concerning the bioactivity of your recombinant produced protein due to unsatisfactory Golgi and ER derived processing and glycosylation?

We are offering the service to produce your protein in an established human cell line. HEK293 (hn embryonic kidney) cells are suitable for the recombinant production of correctly processed and glycosylated proteins especially human proteins. The expressed recombinant proteins show excellent bioactivity compared to molecules produced in E. coli.

The proteins are produced in a serum free culture medium with defined compounds of safe origin. The use of this medium for cultivation guarantees constancy of the production process. Serum free cultivation does justice to the standards for hygiene, abstinence of virus, function and purity of the product.

We offer a complete range of working steps for the recombinant production of your desired protein and deliver the purified protein to you. Cloning of the gene of interest in an appropriate expression vector and preparation of the plasmid DNA is followed by transfection in HEK293 cells and selection of stable clones with high productivity. These are checked for product identity and bioactivity. Selected clones expressing the recombinant protein in secreted form or intracellularly are expanded and adapted to serum-free cultivation in suspension cultures. The expressed protein is purified chromatographically from culture supernatant or cell extract. Purification is carried out to partial or high degree according your individual demands and offered in liquid or lyophilized form.

Lab scale production can be expanded to pilote scale if needed.

Service Description		Price (Euro)
Cloning of GOI	2 weeks	1650,00
Manufacture gene to be expressed		
Cloning of gene into eukaryontic expression vector		
Plasmid purification and sequencing		
Transfection in recipient HEK293 cells		250,00
Transfer of expression construct in HEK293		
Selection of High Producer Clones	6 weeks	3300,00
Isolation of clones with high productivity and stable expression.		
Verification of the Expressed Protein		500,00
Checking of identity of the expressed protein by protein -, analytical-, and immunological screenings.		
Adaptation to Serum-Free Cultivation	4 weeks	1200,00
Gradual adaptation of cells to serum free cultivation in suspension		
Lab Scale Production		500,00 / L
Cultivation of recombinant cells in lab scale and isolation of the product from culture supernatant or cell extract.		
Chromatographic Purification	On time and material base	
Chromatographic purification and concentration of the produced protein		
Checking of Biological Activity	On time and material base	
Carrying out of enzymatic and cell based assays for determination of bioactivity		



Proteins from human cell lines

Recombinant Human Cellular Fibronectin

Description

Fibronectin is a high molecular mass, multidomain glycoprotein which plays a key role in various cell-cell and cell-substrate processes such as cell attachment, spreading, growth, differentiation and migration. It is present at cell surfaces and contributes to ECM architecture. There are binding activities for collagen, heparin, fibrin, bacteria and DNA.

Fibronectin is composed of two subunits each consisting of three types of homologous repeating sequences called type I, II, III homologies. These different structural segments contribute to the versatile functions of the molecule.

Cellular fibronectin compared to plasma FN contains the extra domain IIICS which probably contributes to different biological activity of the protein.

Source

- original DNA sequence for cellular fibronectin
- produced serum free in a human cell line
- purified from the culture supernatant

Purification

Protein purified by affinity and gel filtration chromatography without use of denaturing agents

Formulation

- liquid or freeze dried (sterile)

Purity

>95% SDS-PAGE (HMS 800 kDa, reduced dimer at 400 kDa - monomer at 220 kDa)

Identity

Immuno-Blot with specific polyclonal IgG

Activity

tested in a cell attachment assay

Stability

at least 12 months at -20° C (avoid repeated freeze/thaw cycles when dissolved)

Pack-Sizes

500 µg and 1000 µg

Usage: laboratory reagent - not to be used as a diagnostic product or for administration to humans or used for any drug purposes

Recombinant Human Vitronectin

Description

Vitronectin is a multifunctional glycoprotein present in blood and in the extra-cellular matrix. The complete open reading frame encodes for 459 amino acids which are preceded by a 19 amino acid signal peptide. It contains three glycosylation sites and its carbohydrate moiety contributes about 30% to this molecular mass.

Vitronectin is an important participant in a large variety of biological functions. These include cell attachment, spreading and migration, blood coagulation, plasminogen activation, fibrinolysis, and the regulation of complement function.

Recombinant vitronectin appears as multimer compared to the one-chain form in blood. This altered structure may contribute to different biological activities.

Source

- original DNA sequence for vitronectin
- produced serum free in a human cell line
- purified from the culture supernatant

Purification

by affinity chromatography without the use of denaturing agents

Formulation

- liquid or freeze dried (sterile)

Purity

>95% SDS-PAGE (HMS 800 kDa, reduced dimer at 120 kDa, monomer at 65 kDa)

Identity

Identity confirmed by Immuno-Blot with specific polyclonal antiserum

Activity

tested in a cell attachment and spreading assay

Stability

at least 12 months at -20° C (avoid repeated freeze/thaw cycles when dissolved)

Pack-Sizes

250 µg 500 µg 1000 µg

Literature

Seger D., Shaltiel S. (2000) FEBS Lett. 480, 169-174

Schwartz I. et al. (1999) IJBCB 31, 539-544

Suzuki S. et al. (1985) EMBO J. 4, 2519-2524



Recombinant Human Decorin

Description

Human Decorin, which is synthesized as a prepro form with 359 amino acids, is a member of the family of small chondroitinsulphate/ dermatan sulphate proteoglycans. Its mature form consists of a core protein of 329 amino acids, a single glycosaminoglycan chain bound to serine 4 and either two or three asparagine-bound oligosaccharides. The central part of the core protein consists of 11 repeats of a leucine-rich sequence motif.

Decorin is widely distributed in all kinds of tissues. It binds collagen type I, II, VI and has been observed associated with collagen fibrils. Decorin can retard collagen fibrillogenesis.

Source

- full-length cDNA without tags cloned in a eucaryotic vector
- produced serum free in a human cell line
- purified from the culture supernatant

Purification

by ion exchange chromatography without the use of denaturing agents

Formulation

- liquid or freeze dried (sterile and non-sterile)

Concentration

approx. 1500 µg/ml

Purity

>95% SDS-PAGE, dispersed band centred at 120 kDa

Identity

Immuno-Blot with specific polyclonal IgG

Activity

tested in a collagen type I inhibition assay

GAG/Protein: approx. 1,5 : 1

Stability

at least 12 months at -20° C, avoid repeated freeze/thaw cycles

Pack-Sizes

250 µg 500 µg 1000 µg Bulk

Literature

Kresse, H. et al. (1994) Proteoglycans ed. by P. Jolles Birkhäuser Verlag

Hausser, H. et al. (1998) Arch. Bioch. Biophys. 349, 363-370

Iozzo, R.V. et al. (1998) J. Biol. Chem. 274, 4489-4492

Yamaguchi, Y., Ruoslahti, E. (1988) Nature 336, 244-246



Recombinant Human Thrombospondin 1

Description

Thrombospondin is a large, trimeric, modular glycoprotein composed of three identical subunits with 1170 amino acids. It is a major constituent of platelets released from its alpha-granules in response to thrombin stimulation. TSP is also a transient component of extracellular matrix in developing and repairing tissues. TSP is a multifunctional protein consisting of distinct structural domains. It contains an aminoterminal domain with a heparin-binding site, a segment that is homologous to the alpha1 chain of type 1 procollagen (PC), three type 1 (properdin) repeats, three type 2 (EGF-like) repeats, seven type 3 (Ca-binding) repeats and a carboxy-terminal cell-binding domain. The different segments are assigned to cell attachment, cell movement, modulation of proliferation, platelet aggregation, modulation of angiogenesis, neurite outgrowth and platelet aggregation. TSP is synthesized and secreted by platelets, fibroblasts, endothelial cells, smooth muscle cells and tumour cells. It functions in proliferation of cells, wound healing, angiogenesis and tumorigenesis.

Source

- original DNA sequence for human TSP 1
- produced serum free in a human cell line
- purified from culture supernatant

Purification

by affinity chromatography and gel filtration

Formulation

- liquid or dried -
- sterile and non-sterile -

Purity

>95% SDS-PAGE (non-reduced band at 450 kDa,)

Identity

Identity confirmed by Immuno-Blot with specific monoclonal IgG

Activity

tested in functional and cell adhesion assays

Stability

at least 12 months at -20° C (avoid repeated freeze/thaw cycles)

Pack-Sizes

250 µg 500 µg 1000 µg

Literature

Adams J. C., Tucker, R.P. (2000) Dev Dyn 218, 280-99

Lawler J. (2000) Curr Opin Cell Biol 12, 634-40

Albo D. et al. (2000) Br J Cancer 83, 298-306

Bornstein P. (2000) Matrix Biol 19, 555-6