

DESCRIPTION

Source *E. coli*-derived human Betacellulin/BTC protein
Asp32-Gln118, with an N-terminal Met
Accession # P35070.1
Produced using non-animal reagents in an animal-free laboratory.
Manufactured and tested under cGMP guidelines.

N-terminal Sequence Analysis Met-Asp32-Gly-Asn-Ser-Thr-Arg-Ser-Pro-Glu

Predicted Molecular Mass 9.9 kDa

SPECIFICATIONS

SDS-PAGE 12-15 kDa, under reducing conditions.

Activity Measured in a cell proliferation assay using Balb/3T3 mouse embryonic fibroblast cells. Rubin, J.S. *et al.* (1991) Proc. Natl. Acad. Sci. USA 88:415.
The ED₅₀ for this effect is 0.100-1.50 ng/mL.

Endotoxin Level <0.10 EU per 1 µg of the protein by the LAL method.

Purity >97%, by SDS-PAGE with quantitative densitometry by Coomassie® Blue Staining.

Mass Spectrometry The molecular weight by mass spectrometry is 9884 Da ± 5 Da.

Host Cell Protein <1.00 ng per µg of protein when tested by ELISA.

Mycoplasma Negative for Mycoplasma.

Host Cell DNA <0.0100 ng per µg of protein when tested by PCR.

Formulation Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 100-500 µg/mL in PBS.

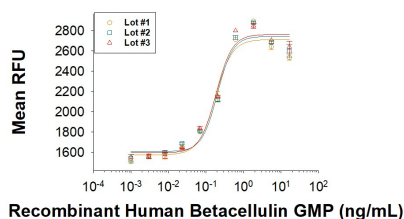
Shipping The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.

Stability & Storage **Use a manual defrost freezer and avoid repeated freeze-thaw cycles.**

- A minimum of 12 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, ≤ -20 °C under sterile conditions after reconstitution.

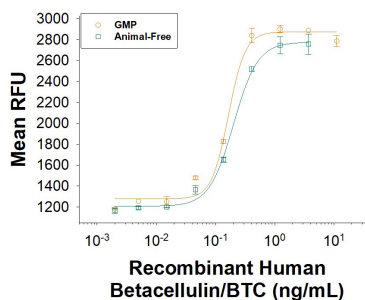
DATA

Bioactivity



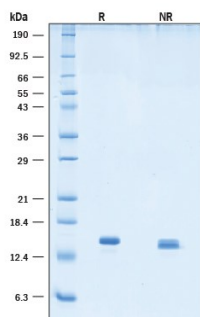
Recombinant Human Betacellulin GMP Protein Bioactivity. GMP-grade Recombinant Human Betacellulin (Catalog # BT-BTC-GMP) as measured in a cell proliferation assay using Balb/3T3 mouse embryonic fibroblast cells. The ED₅₀ for this effect is 0.100-1.50 ng/mL. Three independent lots were tested for activity and plotted on the same graph to show lot-to-lot consistency of GMP Betacellulin.

Bioactivity



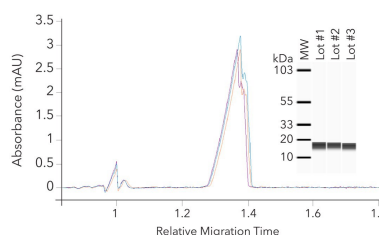
Equivalent Bioactivity of GMP and Animal-Free grades of Recombinant Human Betacellulin/BTC. Equivalent bioactivity of GMP (Catalog # BT-BTC-GMP) and Animal-Free (Catalog # BT-BTC-AFL) grades of Recombinant Human Betacellulin/BTC as measured in a cell proliferation assay using Balb/3T3 mouse embryonic fibroblast cells (orange and green, respectively).

SDS-PAGE



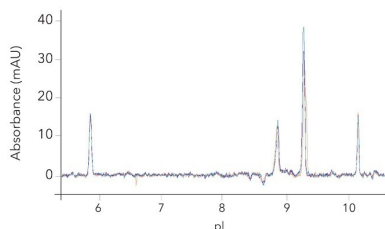
Recombinant Human Betacellulin GMP Protein SDS-PAGE 2 ug/lane of GMP-grade Recombinant Human Betacellulin (Catalog # BT-BTC-GMP) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing bands at 13-15 kDa and 14-15 kDa, respectively.

Data



Recombinant Human Betacellulin GMP Protein Purity Analysis by Size Separation. Three independent lots of Recombinant Human Betacellulin GMP Protein (Catalog # BT-BTC-GMP) were analyzed by Maurice CE-SDS PLUS (IS is an Internal Standard). A gel-like representation of the purity analysis data (inset) can be obtained from the Lane View feature in Compass software for iCE. Profiles from the three runs were superimposed, showing excellent manufacturing consistency.

Data



Recombinant Human Betacellulin GMP Protein Purity Analysis by Charge Variation. Three independent lots of Recombinant Human Betacellulin GMP Protein (Catalog # BT-BTC-GMP) were analyzed by Maurice icIEF using native fluorescence detection (Mkr 5.85 and 9.99 are pI Markers). Profiles from the three runs were superimposed, showing excellent manufacturing consistency.

BACKGROUND

Betacellulin (BTC) is a new member of the EGF family of cytokines that is comprised of at least ten proteins including EGF, TGF- α , amphiregulin, HB-EGF, and the various heregulins. All of these cytokines are synthesized as transmembrane precursors and are characterized by the presence of one or more EGF structural units in their extracellular domain. The soluble forms of these cytokines are released by proteolytic cleavage. BTC, a heparin-binding protein, was originally isolated from the conditioned media of mouse pancreatic beta tumor cells as a 32 kDa glycoprotein composed of 80 amino acid residues. The cDNA encoding human BTC was cloned from a human breast adenocarcinoma cell line (MCF-7) cDNA library. Human and mouse cDNAs encode BTC precursor proteins of 178 and 177 amino acid residues, respectively. At the amino acid sequence level, human BTC precursor protein exhibits 79% identity with that of the mouse BTC precursor. In a mouse cell line transfected with human BTC cDNA, three forms of soluble human BTC have been detected: the glycosylated, intact BTC composed of 80 amino acid residues, a truncated molecule lacking 12 amino acid residues from the amino terminus, and a second truncated molecule lacking 30 amino acid residues from the amino terminus. The biological activities of these BTC forms were shown to be identical. BTC can bind to the EGF receptor and is a potent mitogen for Balb/c 3T3 fibroblasts, retinal pigment epithelial cells and vascular smooth muscle cells.

MANUFACTURING SPECIFICATIONS

GMP Proteins

R&D Systems, a Bio-Techne Brand's GMP proteins are produced according to relevant sections of the following documents: USP Chapter 1043, Ancillary Materials for Cell, Gene and Tissue-Engineered Products and Eu. Ph. 5.2.12, Raw Materials of Biological Origin for the Production of Cell-based and Gene Therapy Medicinal Products.

R&D Systems' quality focus includes:

- Manufactured and tested under an ISO 9001:2015 and ISO 13485:2016 certified quality system
- Documented processes and QA control of documentation and process changes
- Personnel training programs
- Raw material testing and vendor qualification/monitoring
- Fully validated equipment, processes and test methods
- Equipment calibration schedules using a computerized calibration program
- Facility maintenance, safety programs and pest control
- Material review process for variances
- Monitoring of stability over product shelf-life

R&D Systems strives to provide our customers with the analytical characteristics of each product so that customers may determine whether our products are appropriate for their research. The Certificate of Analysis provided contains the following lot specific information:

- N-terminal amino acid analysis, SDS-PAGE analysis, and endotoxin level (as determined by LAL assay) performed on each bulk QC lot, not on individual bottlings of each QC lot
- Post-bottling lot-specific bioassay results (compliance with an established range) and results of microbial testing according to USP <71>
- Host Cell Protein testing performed by ELISA
- Mycoplasma testing by ribosomal RNA hybridization assay

Additional testing and documentation requested by the customer can be arranged at an additional cost.

Production records and facilities are available for examination by appropriate personnel on-site at R&D Systems.

R&D Systems sells GMP grade products for preclinical or clinical *ex vivo* cell therapy applications. They are not for *in vivo* use. Please read the following End User Terms prior to using this product.

Animal-Free Manufacturing Conditions

Our dedicated controlled-access animal-free laboratories ensure that at no point in production are the products exposed to potential contamination by animal components or byproducts. Every stage of manufacturing is conducted in compliance with R&D Systems' stringent Standard Operating Procedures (SOPs). Production and purification procedures use equipment and media that are confirmed animal-free.

Production

- All molecular biology procedures use animal-free media and dedicated labware.
- Dedicated fermentors are utilized in committed animal-free areas.

Purification

- Protein purification columns are animal-free.
- Bulk proteins are filtered using animal-free filters.
- Purified proteins are stored in animal-free containers in a dedicated cold storage room.

Quality Assurance

- Low Endotoxin Level.
- No impairment of biological activity.
- High quality product obtained under stringent conditions.

[Please read our complete Animal-Free Statement.](#)

PRODUCT SPECIFIC NOTICES**END USER TERMS OF USE OF PRODUCT**

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You agree to review these End User Terms of Use of Product to ensure any subsequent use by you of R&D Systems' GMP Products following changes to these End User Terms of Use of Product constitutes your acceptance of all such changes.

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