SibTech, Inc.

ctEGF

SBT201

ctEGF is an engineered 9.3 kDa human epidermal growth factor. The protein consists of human EGF fused to an N-terminal Cys-tag (ct) via a $(G_4S)_3MG$ linker. Complete amino acid sequence of Cys-tagged EGF has the following 86 amino acids:

Met Lys Glu Ser Cys Ala Lys Lys Phe Gln Arg Gln His Met Asp Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Met GlyAsn Ser Asp Ser Glu Cys Pro Leu Ser His Asp Gly Tyr Cys Leu His Asp Gly Val Cys Met Tyr Ile Glu Ala Leu Asp Lys Tyr Ala Cys Asn Cys Val Val Gly Tyr Ile Gly Glu Arg Cys Gln Tyr Arg Asp Leu Lys Trp Trp Glu Leu Arg

Purification: ctEGF is produced in *E. Coli* and purified from inclusion bodies with SH-group of C4 residue "protected" in a mixed disulfide with glutathione. ctEGF is purified by ion-exchange chromatography to >95% purity and lyophilized from 20 mM ammonium bicarbonate. Purified ctEGF migrates as a single band with an apparent molecular weight of 10 kDa in reducing SDS-PAGE.

Functional activity: The ability of ctEGF to bind to EGF receptor is tested *in vitro* on MDA-231luc human breast cancer cells (SibTech product # SBT093.MDA). Relative to recombinant human EGF (Sigma), ctEGF displays 95-100% EGF activity.

Radiolabeling with ^{99m}Tc and other applications: After treatment with equimolar amounts of DTT, C4-thiol group in Cys-tag becomes available for site-specific radiolabeling with ^{99m}Tc for SPECT imaging of EGF receptors, or conjugation of various payloads (1, 2).

One vial contains 0.1 mg of essentially salt-free lyophilized ctEGF

Reconstitution: To insure full recovery, centrifuge the vial briefly before opening. Reconstitute in 0.1 ml of sterile PBS, to a final concentration of 1 mg/ml. We do not recommend using less than 0.1 ml for reconstitution.

Stability: Lyophilized ctEGF is stable for 1 year at -20^{o}C. After reconstitution, ctEGF is stable and functionally active for at least 6 months, if stored at -20^{o}C or below. Multiple thawing-freezing should be avoided.

Safety warnings: For research use only. Not for human use. Not recommended or intended for diagnosis in humans or animals. As all chemicals should be considered as potentially hazardous, it is advisable to wear suitable protective clothing, such as laboratory overalls, safety glasses and gloves. Care should be taken to avoid contact with skin or eyes. In case of contact with skin or eyes, wash immediately with water.

References:

1. Levashova Z, Backer M, Horng G, Felsher D, Backer JM, Blankenberg FG. SPECT and PET Imaging of EGF Receptors with Site-Specifically Labeled EGF and Dimeric EGF. Bioconjug. Chem. *20*, 742–749, 2009.
2. Backer MV, Levashova Z, Levenson R, Blankenberg FG, Backer JM. Cysteine-containing fusion tag for site-specific conjugation of therapeutic and imaging agents to targeting proteins. Methods in Molecular Medicine. Peptide-based Drug Design. Humana Press, New York, NY. Ed: L. Otvos. Vol. 494, p.275-94, 2008.

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