Product Data Sheet

Product Name: RGD-4C

Catalog Number: 29897 (1 mg) Lot Number: See label on vial
29898 (5 mg)

Sequence: H-Ala-Cys-Asp-Cys-Arg-Gly-Asp-Cys-Phe-Cys-Gly-OH
(Disulfide bridge: 2-10 and 4-8) (3-letter code)
ACDCRGDCFCG (Disulfide bridge: 2-10 and 4-8) (1-letter code)

Molecular Weight: 1146.3

% Peak Area by HPLC: ≥ 95

Appearance: Lyophilized white powder

Peptide Reconstitution: RGD-4C peptide is freely soluble in H_2O.

Storage: RGD-4C peptide is shipped at ambient temperature. Upon receipt, store lyophilized peptide at –20°C or lower. Reconstituted peptide can be aliquoted and stored at –20°C or lower.

Description: This peptide, a double cyclic peptide, binds preferentially to integrins at sites of tumor angiogenesis and inflamed synovium *in-vivo*, and can be internalized into targeted cells. Ref: Curnis, F. et al. *Cancer Res*. **64**, 565 (2004); Su, Z. et al. *Bioconjug. Chem*. **13**, 561 (2002); Gerlag, M. et al. *Arthritis Res.* **3**, 357 (2001); Ellerby, H. et al. *Nature Med.* **5**, 1032 (1999).

Additional Information: *Listed below are relevant information that may provide a guideline on how to use this product. End users will have to adapt to their own specific applications.*

RGD4C (KACDCRGDCFCG, molecular weight [MW] 1,273.9) was obtained from AnaSpec Inc. and RGE4C (KACDCRGECFCG, MW 1,288.3) was obtained from Biopolymer Core Facility (University of Maryland, Baltimore, MD). RGD4C or RGE4C was dissolved in dry *N*,*N*-dimethylformamide (DMF), dried over 4-Å molecular sieves). With constant stirring, polymeric precursor in dry DMF and dry pyridine (1:1 molar equivalents relative to the polymeric ONp content) were added to the peptide solution (1.3 times excess molar equivalents relative to ONp). The reaction mixture was bubbled with nitrogen and continuously stirred at room temperature for 22 h. The reaction was terminated with 84 µL of 0.1N sodium hydroxide-Line, B. R. et al. *J. Nucl. Med.* **46**, 1552 (2005).

In selected experiments, exogenous peptides expressing an RGD sequence (RGD-4C; AnaSpec, San Jose, CA) were used as blocking agents to evaluate the role of α_v integrins in mediating viral entry and transduction . BM-DC ($5x10^5$) were preincubated with 20 µM or 200 µM RGD-4C peptide at 4°C for 60 min. Ad vectors (Ad-GFP or Ad-RGD-GFP) were then added at a MOI of 1000 and incubated at 4°C for the first hour, followed by a 37°C incubation for the second hour. Cells were treated with 1 mM trypsin-EDTA (Invitrogen, Calsbad, CA) for 2 min at 37°C and

then DNase (0.02 mg/ml, Sigma Chemical Co.) for 10 min at 37°C to disrupt and degrade vectors, which had not been internalized. After additional washing, cells were incubated for a final 40–48 h prior to assessing transduction efficiency by FACS-Harui, A. et al. *J. Leukocyte Bio.* **79**. 1271 (2006).

RGD4C (KACDCRGDCFCG, M_w 1273.9) was obtained from AnaSpec (San Jose, CA). RGD4C was dissolved in dry *N*,*N*-DMF (dried over 4-Å molecular sieves). Under constant stirring, dry pyridine (1:1 molar equivalents relative to the polymeric ONp content) followed by polymeric precursor in dry DMF was added to the RGD4C solution (1.3 times excess molar equivalents relative to ONp). The reaction mixture was bubbled with nitrogen and continuously stirred at room temperature for 22 h. The reaction was terminated with 54 µl of 1-amino-2-propanol-Mitra, A. et al. *Nucl. Med. & Bio*. **33**, 43 (2006).

The peptides $_D$(KLAKLAK)$_2$, ACDCRGDCFC-GG-$_D$(KLAKLAK)$_2$, and ACDCRGDCFC (RGD-4C) were synthesized to our specifications at higher than 90% purity by high-performance liquid chromatography (Anaspec, San Jose, CA, USA). The identities of the peptides were verified by mass spectrometry. For the retina-targeting experiments, 1 x 10^9 transducing units (TUs) of CPRECES- (displaying the peptide insert CPKVCPRECESNC), NGR- (displaying the peptide insert CNGRC), RGD-4C- (displaying the peptide insert ACDCRGDCFC), or fd-tet-phage in 100 µl of DMEM were injected intravenously through the tail vein into either P18 C57B/6 mice that had been exposed to 75% oxygen from days P7-P12 or into P18 C57B/6 mice not treated with oxygen-Lahdenranta, J. et al. *FASEB* **21**, 3272 (2007).

Published Citations:

Line, B. R. et al. *J. Nucl. Med.* **46**, 1552 (2005).
Mitra, A. et al. *J. Control. Releases* **102**, 191 (2005).
Harui, A. et al. *J. Leukocyte Bio.* **79**. 1271 (2006).
Kolonin, M. G. et al. *Cancer Res.* **66**, 34 (2006).
Mitra, A. et al. *Nucl. Med. & Bio*. **33**, 43 (2006).
Lahdenranta, J. et al. *FASEB* **21**, 3272 (2007).
Wang, H. et al. *Mol. Cancer Ther*. **7**, 1044 (2008).
Dane, KY. et al. *Mol. Cancer Ther.* **8**, 2312 (2009).

Related Products:

Name	Cat #	Size
RGD-targeted Proapoptotic Peptide (ACDCRGDCFC-GG-klaklakklaklak-NH2 (S-S bonded C1-C4 & C2-C3))	62207	1 mg

For Research Use Only