**Proteins** 

# Crosstide

Cat. No.: HY-P0315 CAS No.: 171783-05-4 Molecular Formula:  $C_{48}H_{77}N_{17}O_{17}$ Molecular Weight: 1164.23

Sequence: Gly-Arg-Pro-Arg-Thr-Ser-Ser-Phe-Ala-Glu-Gly

Sequence Shortening: **GRPRTSSFAEG** 

Akt Target:

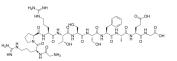
PI3K/Akt/mTOR Pathway:

Storage: Sealed storage, away from moisture

> Powder -80°C 2 years

-20°C 1 year

\* In solvent: -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture)



**Product** Data Sheet

### **SOLVENT & SOLUBILITY**

In Vitro

 $H_2O : \ge 50 \text{ mg/mL } (42.95 \text{ mM})$ 

\* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent Mass Concentration	1 mg	5 mg	10 mg
	1 mM	0.8589 mL	4.2947 mL	8.5894 mL
	5 mM	0.1718 mL	0.8589 mL	1.7179 mL
	10 mM	0.0859 mL	0.4295 mL	0.8589 mL

Please refer to the solubility information to select the appropriate solvent.

#### **BIOLOGICAL ACTIVITY**

Description

Crosstide is a peptide analog of glycogen synthase kinase  $\alpha/\beta$  fusion protein sequence which is a substrate for Akt.

In Vitro

16HBE14o-cells exposed to RV39 demonstrate a crosstide kinase activity in vitro. Serine phosphorylation of crosstide is confirmed by immunoblotting, and phosphorylation is blocked by PP2 but not by PP3<sup>[1]</sup>. The wildtype GST-AKT2 shows significant phosphoryl transferase activity towards crosstide, reaching an initial velocity of 16 pmol phosphate/min/µg enzyme. The mutant GST-AKT2T/E,S/D displayed an initial velocity of 85 pmol phosphate/min/µg kinase for phosphorylation of Crosstide, corresponding to a 5-fold increase compared to the wildtype enzyme<sup>[2]</sup>. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

### **PROTOCOL**

## Kinase Assay [1]

After serum deprivation for 24 h, cells are incubated with digoxigenin-labeled sham protein or digoxigenin-labeled RV39 at an MOI of 1.0 for 10 min. Cell homogenates are immunoprecipitated with mouse anti-digoxigenin antibody and precipitates incubated with Crosstide and [ $\gamma$ -<sup>32</sup>P]ATP. Crosstide is a glycogen synthase kinase  $\alpha/\beta$  fusion protein sequence (GRPRTSSFAEG) which is a substrate for Akt. Samples are processed for autoradiography and immunoblotting using rabbit anti-phospho-Tyr<sup>416</sup> Src, mouse anti-Src (clone GD11), rabbit anti-phospho-Ser<sup>473</sup>, or rabbit anti-Akt. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

#### **CUSTOMER VALIDATION**

• FASEB J. 2021 May;35(5):e21526.

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#### **REFERENCES**

[1]. Bentley JK, et al. Rhinovirus activates interleukin-8 expression via a Src/p110beta phosphatidylinositol 3-kinase/Akt pathway in human airway epithelial cells. J Virol. 2007 Feb;81(3):1186-94. Epub 2006 Nov 22.

[2]. Baer K, et al. Activation of a GST-tagged AKT2/PKBbeta. Biochim Biophys Acta. 2005 Oct 10;1725(3):340-7. Epub 2005 Apr 20.

Caution: Product has not been fully validated for medical applications. For research use only.

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