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## **AKT (Isoforms 1,2,3) Inhibitory Peptide Set**

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<b>Catalog No</b>	<b>IMG-2007</b>
<b>Content:</b>	<b>Akt (Isoforms 1,2,3) Inhibitor peptide: 2 x 1 mg (lyophilized)</b> DRQIKIWFQNRRMKWKK <u>AVTDHPDRLWAWKEF</u> (AKT sequence is underlined). Molecular weight: 4214.  <b>Control peptide: 2 x 1 mg (lyophilized)</b> DRQIKIWFQNRRMKWKK Molecular weight: 2361
<b>Species Reactivity:</b>	Human
<b>Storage:</b>	The solid product is stable in the dessicator at room temperature for year. However, we recommend storing dessicated at -20°C.
<b>Form:</b>	White Solid
<b>Application:</b>	Inhibition of Akt kinase activity.
<b>Inhibitory Mechanism:</b>	Functions as a TCL1 PH decoy by binding to Akt.
<b>Solubility:</b>	Solubilize the peptides prior to use by making 5 mM PBS* stock solutions (please see <b>Preparation of 5 mM Stock Solutions</b> ). The stock solutions are stable at -20°C for 6-8 months. Avoid repeated freeze/thaw cycles. For multiple uses, we suggest aliquoting the stock solution prior to freezing.

### **Background**

Akt is a protein kinase that plays a central role in inhibiting apoptosis through promoting cell survival. Activated Akt functions by phosphorylating downstream targets in survival signaling pathways. TCL1 (a proto-oncogene underlying human T cell prolymphocytic leukemia) interacts with Akt through an N-terminal pleckstrin homology (PH) domain and functions as an Akt kinase co-activator. This inhibitory peptide contains a sequence (AVTDHPDRLWAWKEF) corresponding to the A strand of human TCL1 that interacts with Akt.<sup>1</sup> The peptide binds to the PH domain of Akt and inhibits Akt1, Akt2, and Akt3 kinase activity.

The Akt (Isoforms 1,2,3) inhibitory peptide also contains a protein transduction (PTD) sequence (DRQIKIWFQNRRMKWKK) derived from antennapedia which renders the peptide cell permeable.<sup>2</sup> The control peptide consists of only the PTD sequence.

## **Preparation of 5 mM Stock Solutions**

PBS\* is added directly to the vials to prepare the stock solutions. *Note: Bring the solution to room temperature and quick spin the tubes before opening the caps.*

### **Akt (Isoforms 1,2,3) Inhibitor Peptide: 1 mg of DRQIKIWFQNRRMKWKKAVTDHPDRLWAWKEF**

Add 47.6 ul of PBS\* to the vial to make a 5 mM stock solution. Mix by vortexing. Aliquot and store at -20°C or -80°C. Avoid repeated freeze thawing.

### **Control Peptide: 1 mg of DRQIKIWFQNRRMKWKK**

Add 84.8 ul PBS\* to the vial. Mix by vortexing. Aliquot and store at 20°C or -80°C. Avoid repeated freeze thawing.

### **\*Recipe for 1X PBS:**

1. Dissolve the following in 800ml distilled H<sub>2</sub>O.
  - 8g of NaCl
  - 0.2g of KCl
  - 1.44g of Na<sub>2</sub>HPO<sub>4</sub>
  - 0.24g of KH<sub>2</sub>PO<sub>4</sub>
2. Adjust pH to 7.5 with HCl.
3. Adjust volume to 1L with additional distilled H<sub>2</sub>O.
4. Sterilize by autoclaving

### **Usage:**

The inhibitor peptide is to block Akt1, Akt2, and Akt3 kinase activity. Optimal peptide concentrations and incubation times vary between model systems and should be determined empirically by users. A 100 uM final concentration may be a useful starting point. Please refer to Hiromura et al (2004) for additional information about how the inhibitor peptide has been used to block Ak1, Akt2 and Akt3 kinase activity.

### **Reference:**

1. Hiromura M, F Okada, T Obata, D Auguin, T Shibata, C Roumestand, M Noguchi. Inhibition of Akt kinase activity by a peptide spanning the  $\beta$ A strand of the proto-oncogene TCL1. *J. Biol. Chem.* 279:53407-5341 (2004).
2. Derossi D, AH Joliot, G Chassaings, A Prochiantz. The Third Helix of the Antennapedia Homeodomain Translocates through Biological Membranes. *J Biol Chem.* 269:10444-10450 (1994).