**#6510** Store at -20°C

## SignalSilence® Akt siRNA II

 10 μM in 300 μl (100 Transfections)

rev. 02/11/16



## Species Cross-Reactivity: H, (M, R, C)

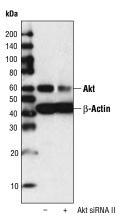
**Description:** SignalSilence<sup>®</sup> Akt siRNA from Cell Signaling Technology allows the researcher to specifically inhibit Akt expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence<sup>®</sup> siRNA products are rigorously tested in-house and have been shown to reduce protein expression by western analysis.

Background: Akt, also referred to as PKB or Rac, plays a critical role in controlling survival and apoptosis (1-3). This protein kinase is activated by insulin and various growth and survival factors to function in a wortmannin-sensitive pathway involving PI3 kinase (2,3). Akt is activated by phospholipid binding and activation loop phosphorylation at Thr308 by PDK1 (4) and by phosphorylation within the carboxy terminus at Ser473. The previously elusive PDK2 responsible for phosphorylation of Akt at Ser473 has been identified as mammalian target of rapamycin (mTor) in a rapamycin-insensitive complex with rictor and Sin1 (5,6). Akt promotes cell survival by inhibiting apoptosis by phosphorylating and inactivating several targets, including Bad (7), forkhead transcription factors (8), c-Raf (9) and caspase-9. PTEN phosphatase is a major negative regulator of the PI3 kinase/Akt signaling pathway (10). LY294002 is a specific PI3 kinase inhibitor (11).

Another essential Akt function is the regulation of glycogen synthesis through phosphorylation and inactivation of GSK-3 $\alpha$  and  $\beta$  (12,13). Akt may also play a role in insulin stimulation of glucose transport (12).

In addition to its role in survival and glycogen synthesis, Akt is involved in cell cycle regulation by preventing GSK-3 $\beta$  mediated phosphorylation and degradation of cyclin D1 (14) and by negatively regulating the cyclin dependent kinase inhibitors p27 Kip (15) and p21 Waf1/CIP1 (16). Akt also plays a critical role in cell growth by directly phosphorylating mTOR in a rapamycin-sensitive complex containing raptor (17). More importantly, Akt phosphorylates and inactivates tuberin (TSC2), an inhibitor of mTOR within the mTOR-raptor complex (18). Inhibition of mTOR stops the protein synthesis machinery by inactivating p70 S6 kinase and activating the eukaryotic initiation factor 4E binding protein 1 (4E-BP1), an inhibitor of translation (18,19).

**Directions for Use:** CST recommends transfection with 100 nM Akt siRNA II 48 to 72 hours prior to cell lysis. For transfection procedure, follow protocol provided by the transfection reagent manufacturer. Please fell free to contact CST with any questions on use.



Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Fluorescein Conjugate) #6201 (-) or SignalSilence® Akt siRNA II (+), using Akt (pan) (11E7) Rabbit mAb #4685 and  $\beta$ -Actin (13E5) Rabbit mAb #4970. Akt (pan) (11E7) Rabbit mAb confirms silencing of Akt expression, while  $\beta$ -Actin (13E5) Rabbit mAb is used to control for loading and specificity of Akt siRNA.

Quality Control: Oligonucleotide synthesis is monitored base by base through trityl analysis to ensure appropriate coupling efficiency. The oligo is subsequently purified by affinity-solid phase extraction. The annealed RNA duplex is further analyzed by mass spectrometry to verify the exact composition of the duplex. Each lot is compared to the previous lot by mass spectrometry to ensure maximum lot-to-lot consistency.

**Specific/Sensitivity:** SignalSilence<sup>®</sup> Akt siRNA II will inhibit human, mouse and rat Akt1 expression. It will not affect Akt2 or Akt3 expression.



**Storage:** Akt siRNA II is supplied in RNAse-free water. Aliquot and store at  $-20^{\circ}$ C.

## Please visit www.cellsignal.com for a complete listing of recommended companion products.

## **Background References:**

(1) Franke, T.F. et al. (1997) Cell 88, 435-7.

Cell Signaling

Orders 877-616-CELL (2355)

Support **S** 877-678-TECH (8324)

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 Applications Key:
 W—Western
 IP—Immunoprecipitation
 IHC—Immunohistochemistry
 ChIP—Chromatin Immunoprecipitation
 IF—Immunofluorescence
 F—Flow cytometry
 E-P—ELISA-Peptide

 Species Cross-Reactivity Key:
 H—human
 M—mouse
 R—rat
 Hm—hamster
 Mk—monkey
 Mi—mink
 C—chicken
 Dm—D. melanogaster
 X—xenopus
 Z—zebrafish
 B—bovine

 Dg—dog
 Pg—pig
 Sc—S. cerevisiae
 Ce-C. elegans
 Hr—Horse
 AII—all species expected
 Species enclosed in parentheses are predicted to react based on 100% homology.