

Datasheet for ABIN4955883

Human and Mouse AKT Pathway Phosphorylation Array C1



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1 Image

1 Publication

Overview

Quantity:	4 samples
Reactivity:	Human, Mouse
Method Type:	Sandwich ELISA
Application:	Antibody Array (AA)

Product Details

Purpose:	C-Series Human and Mouse AKT Pathway Phosphorylation Array C1. Detects 18 phosphorylated human and mouse proteins. Suitable for all liquid sample types but intended for use with cell and tissue lysates.
Sample Type:	Plasma, Cell Culture Supernatant, Serum, Cell Lysate, Tissue Lysate
Analytical Method:	Semi-Quantitative
Detection Method:	Chemiluminescent
Specificity:	Akt (P-Ser473) (PKB (P-Ser473)), AMPKα (P-Thr172), BAD (P-Ser112), 4E-BP1 (P-Thr36), ERK1 (P-T202/Y204)/ERK2 (P-Y185/Y187), GSK3α (P-Ser21), GSK3β (P-Ser9), mTOR (P-Ser2448), p27 (P-Thr198), P53 (P-Ser15), P70S6K (P-Thr421/Ser424), PDK1 (P-S241), PRAS40 (P-Thr246), PTEN (P-Ser380), Raf-1 (Ser301), RPS6 (P-Ser235/Ser236), RSK1 (P-Ser380), RSK2 (P-Ser386)
Characteristics:	<ul style="list-style-type: none"> • Easy to use • No specialized equipment needed • Compatible with nearly any liquid sample • Proven technology (many publications) • Highly sensitive (pg/mL) • Sandwich ELISA specificity • Higher density than ELISA, Western blot or bead-based multiplex

Product Details

Components:	Antibody Array Membranes Biotinylated Detection Antibody Cocktail Blocking Buffer Wash Buffers 1 and 2 Cell & Tissue Lysis Buffer Detection Buffers C and D Plastic Incubation Tray Protease Inhibitor Cocktail (in select kits)
Material not included:	Pipettors, pipet tips and other common lab consumables Orbital shaker or oscillating rocker Tissue Paper, blotting paper or chromatography paper Adhesive tape or Saran Wrap Distilled or de-ionized water A chemiluminescent blot documentation system (such as UVP's ChemiDoc-It® or EpiChem II Benchtop Darkroom), X-ray Film and a suitable film processor, or another chemiluminescent detection system.

Application Details

Application Notes:	<p>Perform ALL incubation and wash steps under gentle rotation or rocking motion (~0.5 to 1 cycle/sec) using an orbital shaker or oscillating rocker to ensure complete and even reagent/sample coverage. Rocking/rotating too vigorously may cause foaming or bubbles to appear on the membrane surface which, should be avoided. All washes and incubations should be performed in the Incubation Tray (ITEM 10) provided in the kit. Cover the Incubation Tray with the lid provided during all incubation steps to avoid evaporation and outside debris contamination. Ensure the membranes are completely covered with sufficient sample or reagent volume during each incubation. Avoid forceful pipetting directly onto the membrane, instead, gently pipette samples and reagents into a corner of each well. Aspirate samples and reagents completely after each step by suctioning off excess liquid with a pipette. Tilting the tray so the liquid moves to a corner and then pipetting is an effective method. Optional overnight incubations may be performed for the following step to increase overall spot signal intensities:</p> <ul style="list-style-type: none">- Sample Incubation- Biotinylated Antibody Cocktail Incubation- HRP-Streptavidin Incubation
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Comment:	The C-Series arrays feature chemiluminescent signal detection. The antibodies are spotted on
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Application Details

nitrocellulose membrane solid supports and are handled in a very similar manner to Western blots.

All C-Series arrays work on the sandwich ELISA principle, utilizing a matched pair of antibodies: an immobilized capture antibody and a corresponding biotinylated detection antibody.

Sample Volume: 1 mL

Plate: Membrane

Protocol:

1. Block membranes
2. Incubate with Sample
3. Incubate with Biotinylated Detection Antibody Cocktail
4. Incubate with HRP-Conjugated Streptavidin
5. Incubate with Detection Buffers
6. Image with chemiluminescent imaging system
7. Perform densitometry and analysis

Restrictions: For Research Use only

Handling

Handling Advice: The antibody printed side of each membrane is marked by a dash (-) or number (#) in the upper left corner. Do not allow membranes to dry out during the experiment or they may become fragile and break OR high and/or uneven background may occur. Grasp membranes by the corners or edges only using forceps. DO NOT touch printed antibody spots.

Storage: -20 °C

Storage Comment: For best results, store the entire kit frozen at -20°C upon arrival. Stored frozen, the kit will be stable for at least 6 months which is the duration of the product warranty period. Once thawed, store array membranes and 1X Blocking Buffer a -20°C and all other reagents undiluted at 4°C for no more than 3 months.

Publications

Product cited in: Platonov, Borovjagin, Kaverina, Xiao, Kadagidze, Lesniak, Baryshnikova, Ulasov: "KISS1 tumor suppressor restricts angiogenesis of breast cancer brain metastases and sensitizes them to oncolytic virotherapy in vitro." in: **Cancer letters**, Vol. 417, pp. 75-88, (2018) ([PubMed](#)).

Image 1.

Western blot analysis of cell lysates from HepG2 cells treated with 100 nM of the indicated inhibitor for 30 min. The blots were probed with the indicated antibodies. The results are representative of three independent experiments.	1	POS	POS	NEG	NEG	Akt (P-Ser473)	AMPK α (P-Thr172)	BAD (P-Ser112)	4E-BP1 (P-Thr36)
	2								
	3	ERK1 (P-T202/Y204)	GSK3 α (P-Ser21)	GSK3 β (P-Ser9)	mTOR (P-Ser2448)	p27 (P-Thr198)	PS3 (P-Ser15)	P70S6K (P-Thr421/Ser424)	PDK1 (P-Ser241)
	4	ERK2 (P-Y185/Y187)							
	5	PRAS40 (P-Thr246)	PTEN (P-Ser380)	Raf-1 (Ser301)	RPS6 (P-Ser235/236)	RSK1 (P-Ser380)	RSK2 (P-Ser386)	NEG	POS
	6								