

Technical Data Sheet

Total Akt2 Flex Set

Product Information

Material Number: 560207
Size: 100 tests
Bead Position: A5
Reactivity: QC Tested: Human
 Reported: Mouse, Rat

Component Description: Total Akt2 PE Detection Reagent
Component Mat. No: 51-9005701
Component Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Component Description: Total Akt2 Capture Bead A5
Component Mat. No: 51-9005702
Component Storage Buffer: Aqueous buffered solution containing fetal bovine serum and ≤0.09% sodium azide.

Component Description: Total Akt2 Standard
Component Mat. No: 51-9005493
Component Storage Buffer: Lyophilized in an aqueous buffered solution containing BSA and ProClin™ 150.

Description

The BD™ CBA Total Akt2 Flex Set is a bead-based immunoassay capable of measuring human, mouse or rat Akt2 (Akt is also known as protein kinase B), a member of the serine/threonine-specific protein kinase family. Human reactivity was determined by testing cell lysates with the BD CBA Total Akt2 Flex Set. The biology and function of Akt2 has been previously reviewed. For more information on bead-based immunoassays, refer to the product insert for the BD CBA Cell Signaling Master Buffer Kit (Cat. No. 560005 or 560006).

Preparation and Storage

This BD™ CBA Flex Set contains one vial of each component listed above. All components of this flex set have been formulated to a 50x concentration to ensure product performance when multiplexed. Store at 4°C. Protect PE Detection Reagent from prolonged exposure to light.

The Total Akt2 Standard provided in this Flex Set is lyophilized and the standard sphere should be transferred to a 1.5 ml microfuge tube for reconstitution. Reconstitute the standard with 0.1 ml Assay Diluent from the BD CBA Cell Signaling Master Buffer Kit (Cat. No. 560005/560006), warm to 37°C and vortex prior to use. After reconstitution, the standard concentration is 50,000 Units/ml and is stable for 3 months when stored at 4°C. When using reconstituted standard, warm to 37°C and vortex to mix thoroughly before use.

Application Notes

Recommended Assay Procedure: The BD CBA Total Akt2 Flex Set must be used in conjunction with a BD CBA Cell Signaling Master Buffer Kit (Cat. No. 560005, 100 tests, or 560006, 500 tests) and a flow cytometer. Detailed instructions on the use of this product can be found in the manual for the BD CBA Cell Signaling Master Buffer Kit. When following the directions in the Master Buffer Kit, the top standard point for the BD CBA Total Akt2 Flex Set will be 1,000 Units/ml. An example standard curve is shown in Figure 1.

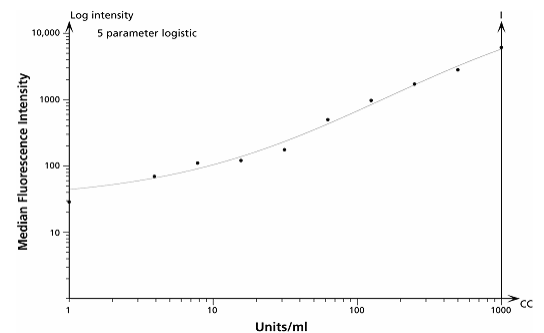


Figure 1. Example BD CBA Total Akt2 Flex Set standard curve. Data acquired on a BD FACSAry bioanalyzer and analyzed using the FCAP Array Software (Cat. No. 641488).

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The BD CBA Total Akt2 Flex Set cannot be used in the same assay well with the following BD CBA Total Flex Set reagents:

Flex Set	Bead Position	Catalog Number
BD CBA Total STAT1 Flex Set	C5	560017
BD CBA Total ZAP70 Flex Set	B8	560019

The BD CBA Total Akt2 Flex Set should not be used in the same assay well with any non-BD CBA Cell Signaling Flex Set reagents (such as BD CBA Human or Mouse Soluble Protein Flex Sets) **nor with any BD CBA Phospho Cell Signaling Flex Set reagents**. For an updated assay compatibility chart for the BD CBA Cell Signaling Flex Sets, please refer to the BD CBA Flex Set System homepage at <http://www.bdbiosciences.com/flexset>.

Performance

Limit of Detection: The theoretical limit of detection is 1.0 Units/ml and was determined by evaluating the estimated result of the average MFI of the negative control (0 Units/ml, $n=30$) + 2 standard deviations.

Specificity		Inter-Assay Reproducibility			Intra-Assay Reproducibility		
		Mean (Units/mL)	Standard Deviation	%CV	Mean (Units/mL)	Standard Deviation	%CV
Total AKT2	Sample 1	30.5	3.1	10%	27.4	4.0	15%
	Sample 2	120.4	13.8	11%	111.7	16.1	14%
	Sample 3	515.0	44.0	9%	492.8	37.9	8%

Reproducibility: The intra-assay and inter-assay reproducibility were determined for the BD CBA Total Akt2 Flex Set by evaluating ten replicates of three different sample levels (intra-assay) and three replicates of three different sample levels from four separate experiments (inter-assay).

Lysate Dilution	Total AKT2	
	Detected (Units/ml)	% of Expected
Neat	1544.1	100%
1 : 2	728.2	94%
1 : 4	374.5	97%
1 : 8	138.4	72%

Linearity: An activated cell lysate was serially diluted to determine the linearity of the assay.

Product Notices

1. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. ProClin is a trademark of Rohm and Haas Company.
4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.