SZABO SCANDIC

Part of Europa Biosite

Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

Weitere Information auf den folgenden Seiten!
See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

linkedin.com/company/szaboscandic

PRODUCT INFORMATION

Cayman LipiDOT Strips™ - PIPs Plus

Item No. 38924

Storage: 2-8°C
Stability: ≥2 years

Information represents the product specifications. Batch specific analytical results are provided on each certificate of analysis.

Description

Cayman LipiDOT Strips™ - PIPs Plus is a nitrocellulose membrane-based screening tool for the identification of protein-lipid interactions and assessment of a protein's relative lipid-binding specificity and affinity. It contains several types of lipids, such as phospholipids, including a variety of phosphatidylinositol phosphate (PIP) species, lysophospholipids, and cardiolipin, as well as cholesterol and sphingolipids. These lipids interact with a variety of proteins to regulate signaling events, nutrient and energy metabolism, lipid transport, and cell adhesion, among other processes.[1]

Cayman LipiDOT Strips™ - PIPs Plus contains 22 biologically relevant lipids and two blank (blue) spots for adding a positive control (protein) or using as a negative control and/or additional lipids in the protein-lipid overlay (PLO) assay. The lipids and controls are spotted on a 3.175 cm x 6 cm supported nitrocellulose membrane in a 3 x 8 grid. This product can be used with Cayman LipiDOT Strips™ - Developing Chamber (Item No. 39380) for the blocking, incubation, and washing steps.

Reagents needed for detecting control protein-lipid interaction:

Reagent	Product Name	Item Number	Description
Control Protein	PtdIns-(4,5)-P_2 (PI(4,5)P_2) Binding Protein (GST-tagged)	10009815	2 µg/ml in blocking buffer
Primary Antibody	GST-tag Polyclonal Antibody	10010013	1:1,500 dilution in blocking buffer
Secondary Antibody	Goat Anti-Rabbit IgG HRP	10004301	1:4,000 dilution in blocking buffer
Wash Buffer			PBS-T (1X PBS, pH 7.2, with 0.1% (v/v) Tween 20)
Blocking Buffer			PBS-T + 3% fatty acid-free BSA

Protocol

Optimized procedure for Cayman LipiDOT Strips™ - PIPs Plus:

1. **Load additional lipid of interest to the membrane (optional):** Spot 1 µl of an additional lipid at 100 µM in a solution, such as chloroform:methanol:water (20:9:1), onto a blue spot on the membrane. Allow the membrane to dry completely in a fume hood.
2. **Load a positive control protein to the membrane (optional):** Spot 0.1-2.0 µg of the protein (1-2 µl), prepared in a suitable aqueous buffer, onto a blue spot on the membrane. Allow the membrane to dry completely in a fume hood.
3. **Load a negative control solution to the membrane (optional):** Spot 1-2 µl of solvent or aqueous buffer alone onto a blue spot on the membrane as a negative control. Allow the membrane to dry completely in a fume hood.
4. **Block:** Cover the Cayman LipiDOT Strips™ membrane in 5-12 ml of freshly prepared blocking buffer overnight at 4°C with gentle agitation.

CAYMAN CHEMICAL
1180 EAST ELLSWORTH RD
ANN ARBOR, MI 48108 · USA
PHONE: [800] 364-9897
[734] 971-3335
FAX: [734] 971-3640
CUSTSERV@CAYMANCHEM.COM
WWW.CAYMANCHEM.COM

PRODUCT INFORMATION

5. **Add the protein of interest:** Discard the blocking buffer and cover the membrane in 5-12 ml of protein solution, at a concentration of 0.5-4 µg/ml, in blocking buffer. Incubate at room temperature for 1 hour with gentle agitation.
6. **Wash:** Discard the protein solution and wash with at least 10 ml of wash buffer 5-6 times, for 10 minutes each, at room temperature with gentle agitation.
7. **Incubate with a primary antibody:** Discard wash buffer and add an appropriate primary antibody prepared in blocking buffer. Incubate at room temperature for 1 hour with gentle agitation.
8. **Wash:** As in Step 6. If a secondary antibody is not needed for detection, skip to Step 11 after this wash.
9. **Incubate with a secondary antibody:** Discard wash buffer and add an appropriate secondary antibody prepared in blocking buffer. Incubate at room temperature for 1 hour with gentle agitation.
10. **Wash:** As in Step 6.
11. **Detect:** Discard wash buffer and detect the bound protein using the detection method of choice.

Suggested Buffers for Optimization:

Wash Buffers	PBS (pH 7.2)
	PBS-T (pH 7.2): 1X PBS, pH 7.2, with 0.1% (v/v) Tween 20
	TBS (pH 7.2): 8 g/L sodium chloride, 0.2 g/L potassium chloride, and 12.1 g/L Tris, pH adjusted
	TBS-T (pH 7.2): TBS, pH 7.2, with 0.1% (v/v) Tween 20
Blocking Buffers	TBS or PBS (pH 7.2) + 3% (w/v) fatty acid-free BSA
	PBS-T (pH 7.2) + 3% (w/v) fatty acid-free BSA
	TBS or PBS (pH 7.2) + 1% (w/v) non-fat dry milk
	TBS or PBS (pH 7.2) + 0.1% (w/v) ovalbumin

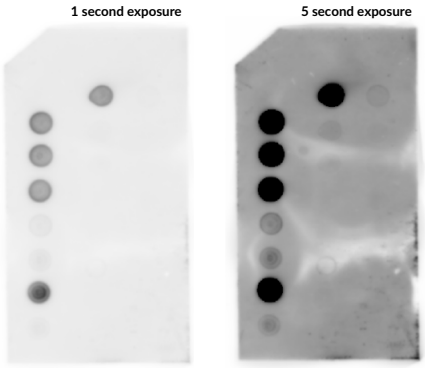
Important Notes:

- The binding of certain proteins may be affected by wash buffer or blocking buffer. The use of different solutions for the protein of interest is recommended in case the optimized protocol does not generate ideal results.
- It is important that the Cayman LipiDOT Strips™ membrane stays wet during all incubation and wash steps. The membrane should be completely submerged in wash/blocking buffer. Use more buffer as needed. Gentle agitation is recommended for all incubation and wash steps.
- The use of freshly prepared wash buffer and blocking buffer is recommended. The blocking buffer should be used within 72 hours of preparation and kept at 4°C when not in use.
- The Cayman LipiDOT Strips™ membrane is designed for purified protein or antibodies only. It is not recommended for cell or tissue lysates.
- Stripping and re-probing the Cayman LipiDOT Strips™ membrane is not recommended.

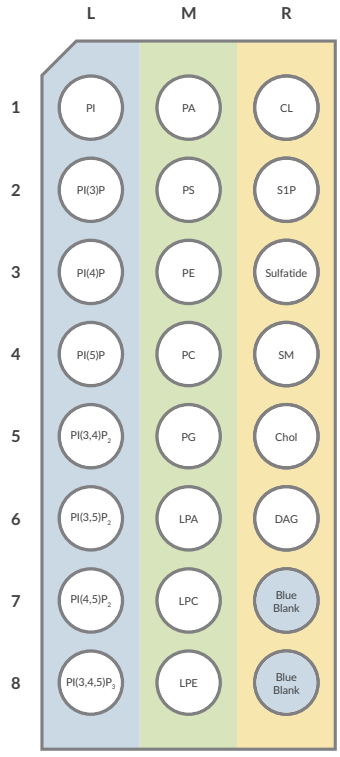
CAYMAN CHEMICAL
1180 EAST ELLSWORTH RD
ANN ARBOR, MI 48108 · USA
PHONE: [800] 364-9897
[734] 971-3335
FAX: [734] 971-3640
CUSTSERV@CAYMANCHEM.COM
WWW.CAYMANCHEM.COM

PRODUCT INFORMATION

Images



The binding of PtdIns-(4,5)-P_2 (PI(4,5)P_2) Binding Protein (GST-tagged) (Item No. 10009815) to Cayman LipiDOT Strips™ - PIPs Plus (Item No. 38924) was detected using the recommended protocol described above.



				M1	Phosphatidic Acid (PA)
				M2	Phosphatidylserine (PS)
				M3	Phosphatidylethanolamine (PE)
				M4	Phosphatidylcholine (PC)
Phosphatidylinositol (PI)	L1	○	○	R1	Cardiolipin (CL)
Phosphatidylinositol-(3)-phosphate (PI(3)P)	L2	○	○	R2	Sphingosine-1-phosphate (S1P)
Phosphatidylinositol-(4)-phosphate (PI(4)P)	L3	○	○	R3	3-Sulfogalactosylceramide (Sulfatide)
Phosphatidylinositol-(5)-phosphate (PI(5)P)	L4	○	○	R4	Sphingomyelin (SM)
Phosphatidylinositol-(3,4)-bisphosphate (PI(3,4)P_2)	L5	○	○	R5	Cholesterol (Chol)
Phosphatidylinositol-(3,5)-bisphosphate (PI(3,5)P_2)	L6	○	○	R6	Diacylglycerol (DAG)
Phosphatidylinositol-(4,5)-bisphosphate (PI(4,5)P_2)	L7	○	○	R7	Blue Blank (Xylene Cyanol FF)
Phosphatidylinositol-(3,4,5)-trisphosphate (PI(3,4,5)P_3)	L8	○	○	R8	Blue Blank (Xylene Cyanol FF)
				M5	Phosphatidylglycerol (PG)
				M6	Lysophosphatidic Acid (LPA)
				M7	Lysophosphatidylcholine (LPC)
				M8	Lysophosphatidylethanolamine (LPE)

Reference

1. Corradi, V., Sejdiu, B.I., Mesa-Galloso, H., *et al.* Emerging diversity in lipid-protein interactions. *Chem. Rev.* **119(9)**, 5775-5848 (2019).

CAYMAN CHEMICAL
1180 EAST ELLSWORTH RD
ANN ARBOR, MI 48108 · USA
PHONE: [800] 364-9897
[734] 971-3335
FAX: [734] 971-3640
CUSTSERV@CAYMANCHEM.COM
WWW.CAYMANCHEM.COM