

TECHNICAL DATA SHEET

Avanti Sphingolipid Snoopers®

Introduction:

Understanding the molecular mechanism behind cellular processes often requires investigating numerous cellular interactions¹. In this regard, many methods have been developed for assaying and identifying unique protein-protein and protein-DNA interactions¹. However, one of the most abundant classes of molecules, lipids, has been widely excluded from such assessments suggesting a need for a screening method that allows for the rapid determination of protein-lipid interactions¹. Avanti Sphingolipid Snoopers® consist of 14 different lipid species spotted individually on a solid support. Each spot contains 1 µg of the highest quality pure lipid and is ideal for investigating lipid-protein interactions.

Brain Sphingomyelin	Lyso SM (d18:1)	Brain Ceramide	Brain Cerebroside	Brain Sulfatide
Total Ganglioside	Brain Sphingosine	So1P	PhytoSo	Cer1P (d18:1/16:0)
Ref.	Sphinganine (d18:0)	Sa1P	DHCer (d18:0/16:0)	DHCer1P (d18:0/16:0)

Figure 1: Avanti Sphingolipid Snooper®

Contents:

Five or ten individually packaged Avanti Snoopers are provided. Each ~3 cm x 6 cm snooper is packaged under argon and contains 1 µg of 14 lipid species. See Figure 1 for the layout of the immobilized lipid species on the Sphingolipid Snooper. For convenience, this template is also found on each individual snooper package.

Storage:

Avanti Snoopers should be stored as packaged at 4°C.

Stability:

Avanti Snoopers are stable for 6 months when stored as packaged at 4°C.

Experimental Protocol:

Materials Needed but Not Provided

- Washing Solution: Tris-Buffered Saline (0.8%

Avanti No.	Description	No. of Strips
330503	Sphingolipid Snoopers®	Packs of 5 & 10 available

NaCl, 20 mM Tris-HCL pH 7.4)

- Blocking Solution: 3% fatty acid free bovine serum albumin (BSA) in Tris-Buffered Saline
- Protein/Antibody Diluent: 1% fatty acid free bovine serum albumin (BSA) in Tris-Buffered Saline (TBS)

General Procedure

- Block: Cover the membrane with ~10 mL of TBS + 3% fatty acid free BSA and incubate for one hour at room temperature with gentle shaking to ensure membrane is completely wetted.*
- Incubate the Snooper with Protein or Antibody of Interest: Remove blocking solution and add desired protein or antibody to the membrane in TBS + 1% fatty acid free BSA for one hour at room temperature with gentle shaking.**
- Wash: Remove protein or antibody solution and wash three times with ~10 mL of TBS for ten minutes with gentle shaking.***
- Secondary Incubation: Following removal of the final wash solution, add a detector or secondary antibody to the membrane in TBS + 3% BSA for one hour at room temperature with gentle shaking.**
- Wash: Repeat wash procedure above to remove any unbound detector or secondary antibody.
- Detection: Following removal of the final wash solution, detect the bound protein or antibody using a method of choice (i.e., colorimetric, chemiluminescence, etc.)

Notes:

In general, it is recommended to approach optimization and troubleshooting as you would with Western blots.

* Ensure that the membrane is completely wetted while blocking, and do not allow the membrane to dry between incubations or washes.

** It is essential to optimize the incubation time and concentration of the protein or antibody of interest, as well as the detector or secondary antibody, in order to reduce background resulting

from non-specific binding.

*** It may be necessary to optimize wash conditions by increasing the number and/or length of washes in order to reduce background signal from non-specific interactions.

Tips:

Wash buffer and diluents containing detergent are not recommended for use with this product, as the stability of individual lipid spots may be compromised.

Avanti Snoopers should not be stripped and re-probed. No data is available concerning the fidelity of lipid species following such treatment.

Reference:

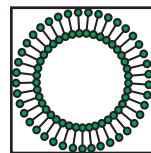
Gallego, O., M.J. Betts, J. Gvozdenovic-Jeremic, K. Maeda, C. Matetzki, C. Aguilar-Gurreri, P. Beltran-Alvarez, S. Bonn, C. Fernandez-Tornero, L.J. Jensen, M. Kuhn, J. Trott, V. Rybin, C.W. Muller, P. Bork, M. Kaksonen, R.B. Russell, and A.C. Gavin. (2010). A systematic screen for protein-lipid interactions in *Saccharomyces cerevisiae*. *Mol Syst Biol* 6:430.

<http://www.ncbi.nlm.nih.gov/pubmed/21119626>

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.



For more details: www.avantilipids.com
Phone: 800-227-0651
Email: lipidomics@avantilipids.com



Avanti[®]
POLAR LIPIDS, INC.