Advanced ImmunoChemical Inc.
Providing for the Advancement of Medical Science

ASSAY NOTES
Product Information

Courtesy of HyTest, Ltd

Gangliosides

Gangliosides are a large group of sialylated glycosphingolipids that are widely expressed in mammalian tissues. Gangliosides are found in most tissues of the body, but they are particularly abundant in brain and nervous tissues. The differential distribution of gangliosides in various tissues is a strong indication that they play important roles in specific functions in different tissues.

Glycosphingolipids (neutral glycosphingolipids and gangliosides) are formed biosynthetically within the Golgi apparatus. Gangliosides are involved in a number of interaction processes with cell external ligands and cell membrane components. Gangliosides seem to be involved in cell-to-cell interaction and regulation of cell signaling. They can be receptors of proteins, viruses and bacteria (GM1 is a receptor of Cholera Toxin). Gangliosides are also playing a role in the cell proliferation. The differentiated cells in human melanoma are expressing GD3 and other b series gangliosides such as GQ1b, whereas GM3 can be involved in the differentiation of some lymphocytic cells.

Applications

Characterization of different cell type-specific disorders can be done using our ganglioside preparations. Incubation of cells in the presence of purified gangliosides leads to insertion of these glycolipids into the cell membranes, specifically altering the binding capacity of the membrane for hormones, bacterial toxins and growth factor.

Single-cell morphology as well as cell-cell interaction and differentiation can be studied by using ganglioside in vivo models. Immunization of animals with purified gangliosides produces anti-ganglioside antibodies. Animals injected with these antibodies are excellent models for studies of epilepsy and other neurological disorders. Removal of sialic acid from purified gangliosides leads to highly potent antigens – asialoglycolipids, which have been shown to be specific determinants of the immune system (B- and T-cell marker). In addition purified gangliosides can be used as biological substrates and inhibitors of gly-cosyltransferases and glycosidases in the study of the metabolic pathway of glycostructures.

High pure gangliosides can be used for characterization of different cell type-specific disorders.

Product Information

Advanced ImmunoChemical offers a number of gangliosides belonging to ganglio-series. The main sources for gangliosides are bovine and human brains. Human gangliosides contain only N-acetylneuraminic acid residues, whereas bovine gangliosides may contain both N-acetyl- and N-glycosylneuraminic acid residues. The bovine brain gangliosides contain the residues of two main sphingosine bases, C18:1 and C20:1 in the ratio 3:2. The final HPLC-purified gangliosides have purity around 98 %. The purity is determined by TLC. Two solvent systems for TLC of gangliosides: the neutral, chloroform – methanol – 15 mM aqueous CaCl2, 60:40:9 (v/v/v) and the basic, chloroform – methanol – 2.5 N aqueous NH3, 60:40:9 (v/v/v) were used. In both systems each ganglioside must be presented by one band (see figures 1 – 5).

The gangliosides are supplied in lyophilized form and they are stable for at least two years at – 20 °C. Almost all gangliosides are soluble in the mixture of chloroform-methanol, 2:1 (v/v). In case of polar gangliosides it can be helpful to increase the amount of methanol or to use pure methanol. Polar gangliosides form micelles in water solution with the critical micelle concentration around 10-7 – 10-8 M. After sterilization by filtration aqueous ganglioside solutions do not require addition of preservative.

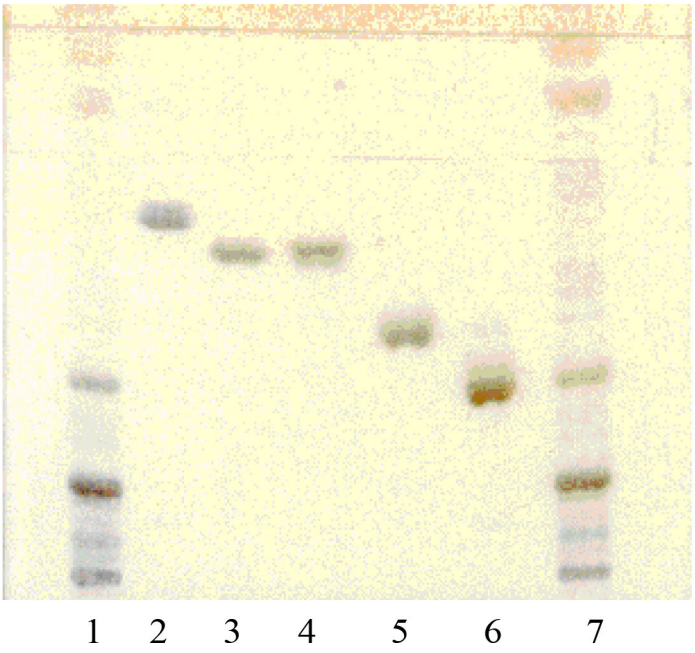

Figure 1. HPTLC of monosialogangliosides in the chloroform – methanol – 15 mM aqueous CaCl2, 60:40:9 (v/v/v), Kieselgel 60 (Merck).

Lines 1 and 7 – mixture of brain glycolipids

Line 2 – GM4 from human brain

Line 3 – GM3 from human brain

Line 4 – GM3 from human liver

Line 5 – GM2 from human brain

Line 6 – GM1 from human brain

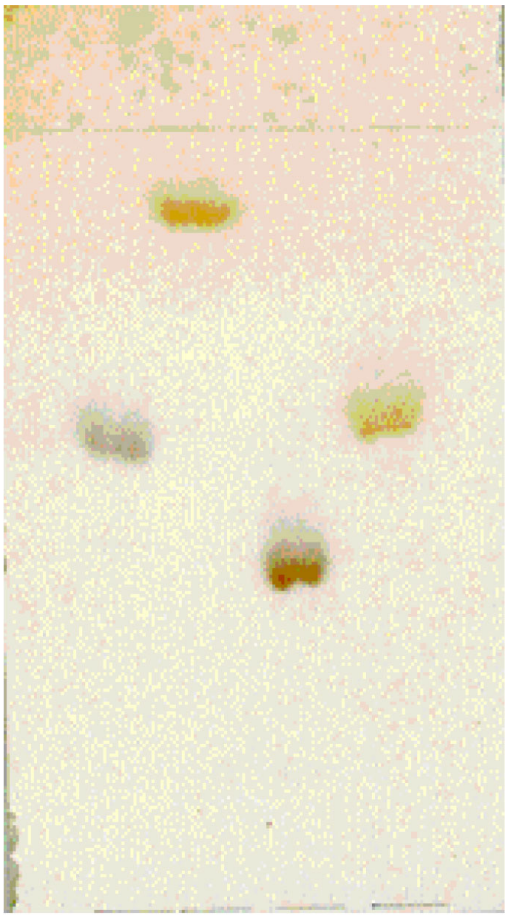

Figure 2. HPTLC of asialo- and monosialogangliosides in the chloroform – methanol – 15 mM aqueous CaCl2, 60:40:9 (v/v/v), Kieselgel 60 (Merck).

Line 1 – GM2 from human brain

Line 2 – Asialo-GM2 from human brain

Line 3 – GM1 from human brain

Line 4 – Asialo-GM1 from human brain

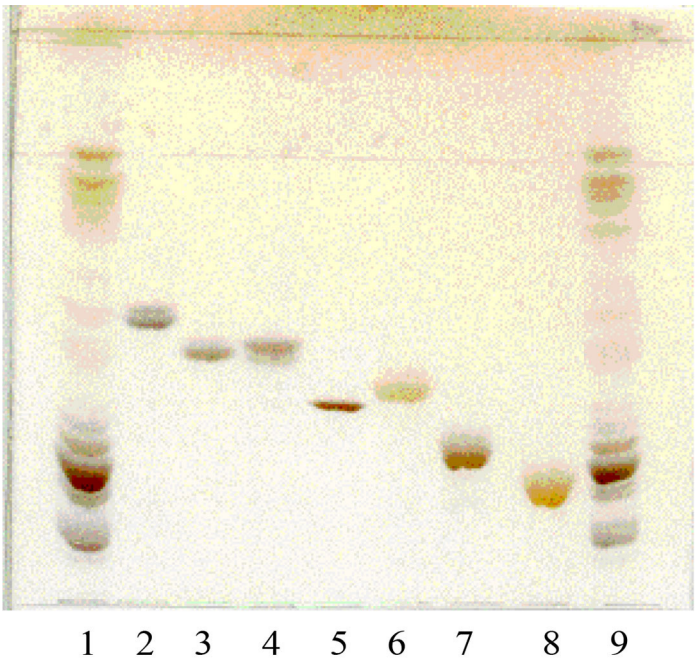

Figure 3. HPTLC of monosialo- and asialogangliosides in the chloroform – methanol – 2.5 N aqueous NH3, 60:40:9 (v/v/v).

Lines 1 and 9 – mixture of brain glycolipids

Line 2 – GM4 from human brain

Line 3 – GM3 from human brain

Line 4 – GM3 from human liver

Line 5 – GM2 from human brain

Line 6 – Asialo-GM2 from human brain

Line 7 – GM1 from human brain

Line 8 – Asialo-GM1 from human brain

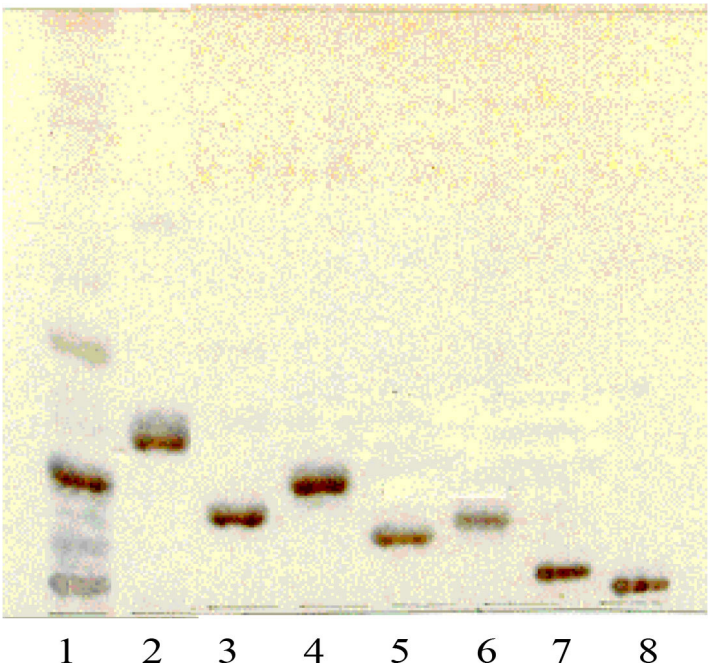

Figure 4. HPTLC of di- and polysialogangliosides in the chloroform – methanol – 15 mM aqueous CaCl2, 60:40:9 (v/v/v)

Line 1 – mixture of brain gangliosides

Line 2 – GD3 from human brain

Line 3 – GD2 from human brain

Line 4 – GD1a from human brain

Line 5 – GD1b from human brain

Line 6 – GT1a from human brain

Line 7 – GT1b from human brain

Line 8 – GQ1b from human brain

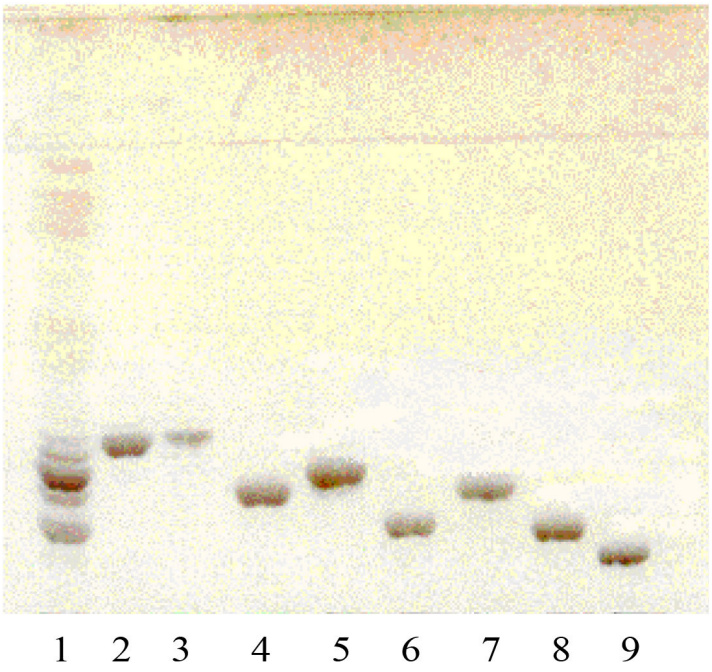

Figure 5. HPTLC of the same gangliosides as on Fig. 4, but in the chloroform – methanol – 2.5 N aqueous NH3, 60:40:9 (v/v/v).

References:

1. Kolter T, et al. (2002) Combinatorial Ganglioside Biosynthesis. J Biol Chem 2. 277(29), 25859-25862**.**

2. Jeyakumar M, et al. (2002) Glycosphingolipid lysosomal storage diseases: therapy and pathogenesis. Neuropathol Appl Neurobiol 28(5), 343-357.

Ordering Information:

Product	Cat #	Purity	Source
Asialoganglioside GM1, Bovine	9-IG4-b	>98%	Bovine Brain MW 1263
Asialoganglioside GM1, Human	9-IG4-h	>98%	Human Brain MW 1263
Asialoganglioside GM2, Bovine	9-IG22-b	>98%	Bovine Brain MW 1103
Asialoganglioside GM2, Human	9-IG22-h	>98%	Human Brain MW 1103
Disialoganglioside GD1a, Bovine	9-IG10-b	>98%	Bovine Brain MW 1827
Disialoganglioside GD1a, Human	9-IG10-h	>98%	Human Brain MW 1827
Disialoganglioside GD1a-NAcGal, Bovine	9-IG10-b-NA	>98%	Bovine Brain MW 2030
Disialoganglioside GD1a-NAcGal, Human	9-IG10-h-NA	>98%	Human Brain MW 2014
Disialoganglioside GD1b, Bovine	9-IG11-b	>98%	Bovine Brain MW 1827
Disialoganglioside GD1b, Human	9-IG11-h	>98%	Human Brain MW 1811
Disialoganglioside GD2, Bovine	9-IG6-b	>98%	Bovine Brain MW 1665
Disialoganglioside GD2, Human	9-IG6-h	>98%	Human Brain MW 1649
Disialoganglioside GD3, Bovine	9-IG7-b	>98%	Bovine Brain MW 1461
Disialoganglioside GD3, Human	9-IG7-h	>98%	Human Brain MW 1438
Monosialoganglioside GM1, Bovine	9-IG1	>98%	Bovine Brain MW 1545
Monosialoganglioside GM1, Human	9-IG1-h	>98%	Human Brain MW 1537
Monosialoganglioside GM2, Bovine	9-IG5-b	>98%	Bovine Brain MW 1383
Monosialoganglioside GM2, Human	9-IG5-h	>98%	Human Brain MW 1375
Monosialoganglioside GM3, Bovine	9-IG8-b	>98%	Bovine Brain MW 1179
Monosialoganglioside GM3, Human	9-IG8-h	>98%	Human Brain MW 1171
Monosialoganglioside GM3, Human (liver)	9-IG9-h	>98%	Human Liver MW 1214
Monosialoganglioside GM4, Bovine	9-IG2-b	>98%	Bovine Brain MW 1017
Monosialoganglioside GM4, Human	9-IG2-h	>98%	Human Brain MW 1009
Tetrasialoganglioside GQ1b, Bovine	9-IG14-b	>98%	Bovine Brain MW 2391
Tetrasialoganglioside GQ1b, Human	9-IG14-h	>98%	Human Brain MW 2359
Trisialoganglioside GT1a, Bovine	9-IG16-b	>98%	Bovine Brain MW 2109
Trisialoganglioside GT1a, Human	9-IG16-h	>98%	Human Brain MW 2085
Trisialoganglioside GT1b, Bovine	9-IG15-b	>98%	Bovine Brain MW 2109
Trisialoganglioside GT1b, Human	9-IG15-h	>98%	Human Brain MW 2085
Globoside, Human	9-IG20-h		Human Erythrocytes MW 1260, The major glycosphingolipid of Human Red Blood Cells
Ganglioside GM1 sphinganine , Bovine	9-IG19-b		Bovine Brain MW 1548, Contains C18:0, C20:0 sphingosine only and saturated fatty acid residues (mostly palmitoyl)

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