3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

Lectin
from *Phaseolus vulgaris*
Erythrohemagglutinin PHA-E

Product Number **L 8629**
Storage Temperature 2-8 °C

Product Description
PHA-E has an approximate MW of 128 kDa.
This product is affinity purified to remove additional protein content. Purity is determined by SDS-PAGE analysis. This product is extensively dialyzed against water prior to lyophilization to render it essentially salt free.

Lectin PHA-E is specifically reactive against erythrocytes. This product is essentially pure isolectin E4.

Lectins are proteins or glycoproteins of non-immune origin that agglutinate cells and/or precipitate complex carbohydrates. Lectins are capable of binding glycoproteins even in presence of various detergents.[1] The agglutination activity of these highly specific carbohydrate-binding molecules is usually inhibited by a simple monosaccharide, but for some lectins, di, tri, and even polysaccharides are required. Lectin PHA-E has not been proven to be specific for any particular oligosaccharide. Lectin PHA-E can promote agglutination of human erythrocytes at 8 µg/ml, but its activity is not inhibited by any monosaccharide such as lactose, galactose, glucose, mannose, etc.[2,3]

Lectins are isolated from a wide variety of natural sources, including seeds, plant roots and bark, fungi, bacteria, seaweed and sponges, mollusks, fish eggs, body fluids of invertebrates and lower vertebrates, and from mammalian cell membranes. The precise physiological role of lectins in nature is still unknown, but they have proved to be very valuable in a wide variety of applications *in vitro*, including:

1. blood grouping and erythrocyte polyagglutination studies.
2. mitogenic stimulation of lymphocytes.
3. lymphocyte subpopulation studies.
4. fractionation of cells and other particles.
5. histochemical studies of normal and pathological conditions.

Sigma offers a range of lectins suitable for the above applications. Most Sigma lectins are highly purified by affinity chromatography, but some are offered as purified or partially purified lectins, suitable for specific applications.

Many of the lectins are available conjugated to (conjugation does not alter the specificity of the lectin):

1. fluorochromes (for detection by fluorimetry).
2. enzymes (for enzyme-linked assays).
3. insoluble matrices (for use as affinity media).

Please refer to the table for general information on the most common lectins.

Procedure
This product has been tested for agglutination using washed red blood cells.
For washing the cells:
1. Transfer whole blood to graduated conical tubes. Use between 5-10 ml of whole blood.
2. Dilute the blood to 10 ml with phospate buffered saline, pH 6.8.
3. Centrifuge the sample at medium speed for 5 minutes in a clinical centrifuge to force the red blood cells to the bottom.
4. Gently remove tha plasma/buffer layer with a pipette.
5. Repeat steps 2-4 three times or until the upper layer is clear.
6. Measure the volume that the red blood cells occupy in the bottom of the tube. Dilute the cells 25-fold with phosphate buffered saline, pH 6.8.

The Agglutination Test is performed as follows:
1. Serially dilute the lectin as follows. Put 50 µl of phosphate buffered saline, pH 6.8 in each of the wells to be used. Add 50 µl of a 1 mg/ml lectin solution to the first well. Remove 50 µl of diluted lectin from the first well and add to the buffer in the next well. Repeat the serial dilutions as desired.

2. Carefully layer 50 µl of washed red blood cells on top of the 50 µl lectin solution.
3. When all the cell suspensions have been added, agitate to mix.
4. Allow to sit for 1 hour at room temperature and then read the results.

Precautions and Disclaimer
For Laboratory Use Only. Not for drug, household or other uses.

Preparation Instructions
This lectin is soluble in phosphate buffered saline, pH 6.8 (1 mg/ml).

Storage/Stability
Aggregation is thought to occur in the presence of high concentrations of 2-mercaptoethanol.

Lectin	MW (kDa)	Subunits	Specificity		Mitogenic Activity
			Blood Group	Sugar	
Abrus precatorius			–		+
Agglutinin	134	4		gal	
Abrin A (toxin)	60	2		gal	
Abrin B (toxin)	63.8	2(αβ)		gal	
Agarius bisporus	58.5	–	–	β-gal(1→3)galNAc	
Anguilla anguilla	40	2	H	α-L-Fuc	
Arachis hypogaea	120	4	T	β-gal(1→3)galNAc	
Artocarpus integrifolia	42	4	T	α-gal→OMe	+
Bandeiraea simplicifolia					
BS-I	114	4	A, B	α-gal, α-galNAc	
BS-I-A_4	114	4	A	α-galNAc	
BS-I-B_4	114	4	B	α-gal	
BS-II	113	4	acq, B, Tk, T	glcNAc	
Bauhinia purpurea	195	4	–	β-gal(1→3)galNAc	+
Caragana arborescens	60; 120[a]	2/4	–	galNAc	
Cicer arietinum	44	2	–	fetuin	
Codium fragile	60	4	–	galNAc	
Concanavalin A	102	4	–	α-man, α-glc	+
Succinyl-Concanavalin A	51	2	–	α-man, α-glc	+[b]
Cytisus scoparius	–	–	–	galNAc, gal	
Datura stramonium	86	2(αβ)	–	$(glcNAc)_2$	
Dolichos biflorus	140	4	A_1	α-galNAc	
Erythrina corallodendron	60	2	–	β-gal(1→4)glcNAc	+
Erythrina cristagalli	56.8	2(αβ)	–	β-gal(1→4)glcNAc	
Euonymus europaeus	166	4(αβ)	B, H	α-gal(1→3)gal	+
Galanthus nivalis	52	4	(h)	non-reduc. α-man	
Glycine max	110	4	–	galNAc	+[c]
Helix aspersa	79	–	A	galNAc	
Helix pomatia	79	6	A	galNAc	
Lathyrus odoratus	40-43	4(αβ)	–	α-man	+
Lens culinaris	49	2	–	α-man	+
Limulus polyphemus	400	18	–	NeuNAc	
Bacterial agglutinin	–	–	–	galNAc, glcNAc	
Lycopersicon esculentum	71	–	–	$(glcNAc)_3$	
Maackia amurensis	130	2(αβ)	O	sialic acid	+
Maclura pomifera	40-43	2(αβ)	–	α-gal, α-galNAc	
Momordica charantia	115-129	4(αβ)	–	gal, galNAc	
Naja mocambique mocambique	–	–	–	–	
Naja naja kaouthia	–	–	–	–	
Narcissus pseudonarcissus	26	2	(h)	α-D-man	
Perseau americana	–	–	–	–	
Phaseolus coccineus	112	4	–	–	
Phaseolus limensis	247(II)	8	A	galNAc	+
	124(III)	4			
Phaseolus vulgaris					
PHA-E	128	4	–	oligosaccharide	+
PHA-L	128	4	–	oligosaccharide	+
PHA-P					
PHA-M					

- Table continued on next page -

Lectin	MW (kDa)	Subunits	Specificity		Mitogenic Activity
			Blood Group	Sugar	
Phytolacca americana	32	–	–	$(glcNAc)_3$	+
Pisum sativum	49	4(αβ)	–	α-man	+
Pseudomonas aeruginosa PA-I	13-13.7	–	–	gal	+[c]
Psophocarpus tetragonolobus	35	1	–	galNAc, gal	
Ptilota plumosa	65; 170	–	B	α-gal	
Ricinus communis					
Toxin, RCA_{60}	60	2	–	galNAc, β-gal	
Toxin, RCA_{120}	120	4	–	β-gal	
Sambucus nigra	140	4(αβ)	–	αNeuNAC(2→6)gal galNAc	+[c]
Solanum tuberosum	50; 100[a]	1, 2	–	$(glcNAc)_3$	
Sophora japonica	133	4	A, B	β-galNAc	
Tetragonolobus purpureas	120(A)	4	H	α-L-fuc	
	58(BA)	2	H	α-L-fuc	
	117(C)	4	H	α-L-fuc	
Triticum vulgaris	36	2	–	$(glcNAc)_2$, NeuNAc	+
Ulex europaeus					
UEA I	68	–	H	α-L-fuc	
UEA II	68	–	–	$(glcNAc)_2$	
Vicia faba	50	4(αβ)	–	man, glc	+
Vicia sativa	40	4(αβ)	–	glc, man	+
Vicia villosa	139	4	$A_{1+}T_n$	galNAc	
A_4	134	4	A_1	galNAc	
B_4	143	4	T_n	galNAc	
Vigna radiata	160	4	–	α-gal	
Viscum album	115	4(αβ)	–	β-gal	
Wisteria floribunda	68	2	–	galNAc	

[a] Concentration-dependent molecular weight
[b] Non-agglutinating and mitogenic
[c] Mitogenic for neuraminidase-treated lymphocytes

References

1. Rueben, L., et al., Activities of lectins and their immobilized derivatives in detergent solutions. Implications on the use of lectin affinity chromatography for the purification of membrane glycoproteins. Biochemistry, **16**, 1787-1794 (1977).
2. Chripeels, M. J. and Raikhel, N.V. Lectin Reviews, vol. 1, pp. 183-197, (New York 1992).
3. Schumacher, U., et al., Lectin Reviews, vol. 1, pp. 195-201, (New York 1992).

IRB/MWM/JRC/NSB/SAG 6/03