

Art no: 05-0038

# CERTIFICATE OF ANALYSIS

Lot no: 291912



Product name:	<b>PHA-P Lectin</b>
Production date:	2023-05
Date of release:	2023-07-19
Stability:	2028-05
Source:	<i>Phaseolus Vulgaris</i> beans (Red Kidney Beans)
Form:	Lyophilized
Storage:	-20 °C (may be shipped at ambient temp)

Analysis	Specification	Result
Appearance	White powder or flocculate	Fulfills requirement
Solubility	1 mg/ml (PBS) within 5 minutes	Fulfills requirement
Assay (%)	≥ 85% (A280) ( $\epsilon$ 1mg/ml = 1.14) in PBS pH 7.4 after filtration through 0.2 $\mu$ m acetate membrane	85.7%, fulfills requirement
Electrophoresis	One major band in SDS-PAGE, comparable to reference sample	Fulfills requirement, see appendix 1
Bacterial burden	Less than 100 CFU per 1 g of solid substance	Fulfills requirement
Haemagglutination	Agglutinates a 2% suspension of human blood group 0 erythrocytes at a lectin concentration $\leq$ 8 $\mu$ g/ml	2.0 $\mu$ g/ml, fulfills requirement, see appendix 2
Ion exchange peak separation	Separation of iso-lectin peaks $\geq$ 1.20 between L3E1 and L2E2, and $\geq$ 1.45 between L2E2 and L1E3	1.88 between L3E1 and L2E2 1.67 between L2E2 and L1E3 Fulfills requirement

Appendix:  
1. SDS-PAGE  
2. Haemagglutination

The above material has met all quality specifications and has been reviewed by a quality representative.

2023-07-19

Quality Assurance, Anna Eliasson

Date



## Appendix 1: SDS-PAGE

### SDS PAGE, analysis of PHA-P lot 291912

Electrophoresis with Pharmacia Phastsystem (Amersham Biosciences)

#### Material

Phast gel gradient 8 – 25  
Phast gel SDS buffer strips

#### Method

The proteins were dissolved to 2 mg/ml 1:1 in PBS and loading buffer (10 mM TRIS/HCl, 1 mM EDTA, 2.5 % SDS, 50 mM DTT).

- LMW marker was from LMW SDS calibration kit for SDS electrophoresis (GE Healthcare).

MW of proteins included in LMW (14 000 Da - 97 000 Da) marker:

Phosphorylase b	97 000
Albumin	66 000
Ovalbumin	45 000
Carbonic anhydrase	30 000
Trypsin inhibitor	20 100
$\alpha$ -Lactalbumin	14 400

The samples were heated for 5 min. and approx. 1  $\mu$ l was applied on Phast gel (gradient 8 – 25).  
Program; 300v, 7.5 mA, 2.0 W, 80 Vh.

The gels were stained with Coomassie blue for 30 min and then destained.

#### Result



Lane 1	empty
Lane 2	LMW
Lane 3,4	PHA-P lot 291912
Lane 5	LMW
Lane 6,7	PHA-P lot 240804 (reference)
Lane 8	empty

Analysis performed by

*Cynthia Hassinen*  
Cynthia Hassinen 2023-07-03



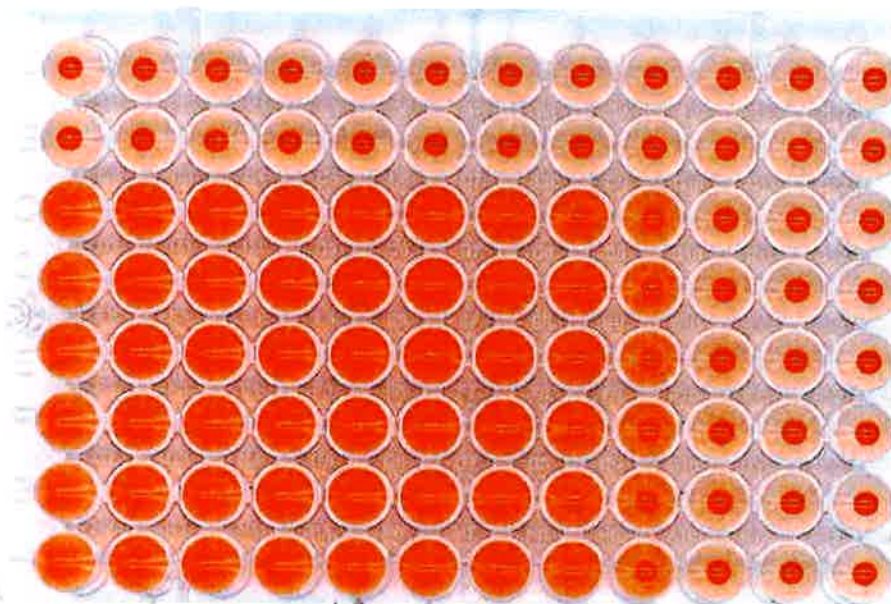
Appendix 2: Haemagglutination

**Haemagglutination (human blood group 0)**

**Lectin from Phaseolus vulgaris (red kidney bean),**

**Phytohemagglutinin (PHA)-P art nr. 05-0038 lot 291912**

Column 1	2	3	4	5	6	7	8	9	10	11	12	
Titer	0	2	4	8	16	32	64	128	256	512	1024	2048
Conc	500	250	125	63	31	16	7.8	3.9	2.0	1.0	0.5	0.2 µg/ml



Row A,B      Blank  
 Row C-H     PHA-P lot 291912

Date/Sign: Cynthia Hassinen  
 Cynthia Hassinen / 2023-06-26