

Art no: 05-0104

# CERTIFICATE OF ANALYSIS

Lot no: 221501



Product name:	<b><i>Lens culinaris</i> lectin, <i>Lens culinaris</i> agglutinin</b>
Production date:	2016-04
Date of release:	2016-04-22
Stability:	2021-04
Form:	Lyophilized

Analysis	Specification	Result
Appearance	White powder or flocculate by visual inspection resulting in a clear solution.	Fulfills requirement
Solubility	Dissolves in distilled water and 0.9% NaCl within 5 minutes.	Fulfills requirement
Assay (%)	> 85 % protein by OD <sub>280</sub> nm ( $\epsilon$ 1mg/ml = 1,34), essentially salt free.	85 %, fulfills requirement
Electrophoresis	Gives two major bands in isoelectrophoresis corresponding to the two isomers LCA-A and LCA-B.	Fulfills requirement, see attachment 3.
Activity Haemagglutination/inhibition	Agglutinates human erythrocytes (2% blood solution) when lectin concentration is $\leq$ 10 $\mu$ g/ml in 0.9 % NaCl after 1 hour at 25 °C. Total inhibition appears with 60 mM Methyl mannoside (end conc. 20 mM) at a titer minimum 4 steps lower than titer of control. Control must have a titer of minimum 32.	Titer inhibition with 20 mM Methyl mannoside = 1; Agglutination at 10 $\mu$ g/ml.  Fulfills requirement, see attachment 2.
UV scan	Typical appearance of <i>Lens culinaris</i> lectin at 700-230 nm.	Fulfills requirement, see attachment 1.
Bacterial burden	Less than 10 <sup>2</sup> CFU per 1 g of solid substance.	< 100 CFU/g, fulfills requirement

<p>Appendixes:</p> <ol style="list-style-type: none"> <li>1. UV Scan 700-230nm.</li> <li>2. Activity, Haemagglutination control.</li> <li>3. IEF electrophoresis.</li> </ol>
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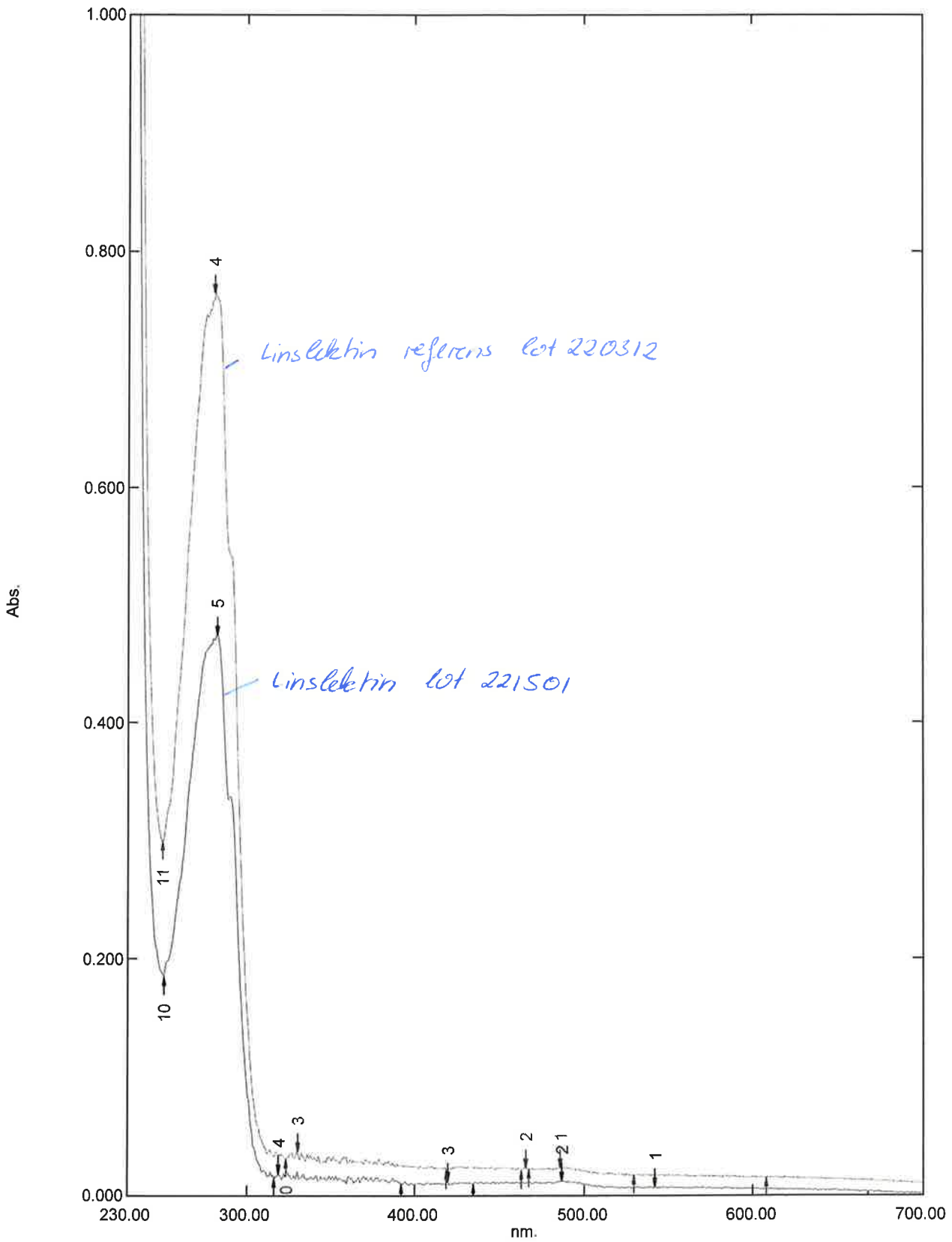
The above material has met all quality specifications and has been reviewed by a quality representative.

  
 Quality Assurance, Erik Silfverplatz

2016-04-22  
 Date

# Overlay Spectrum Graph Report

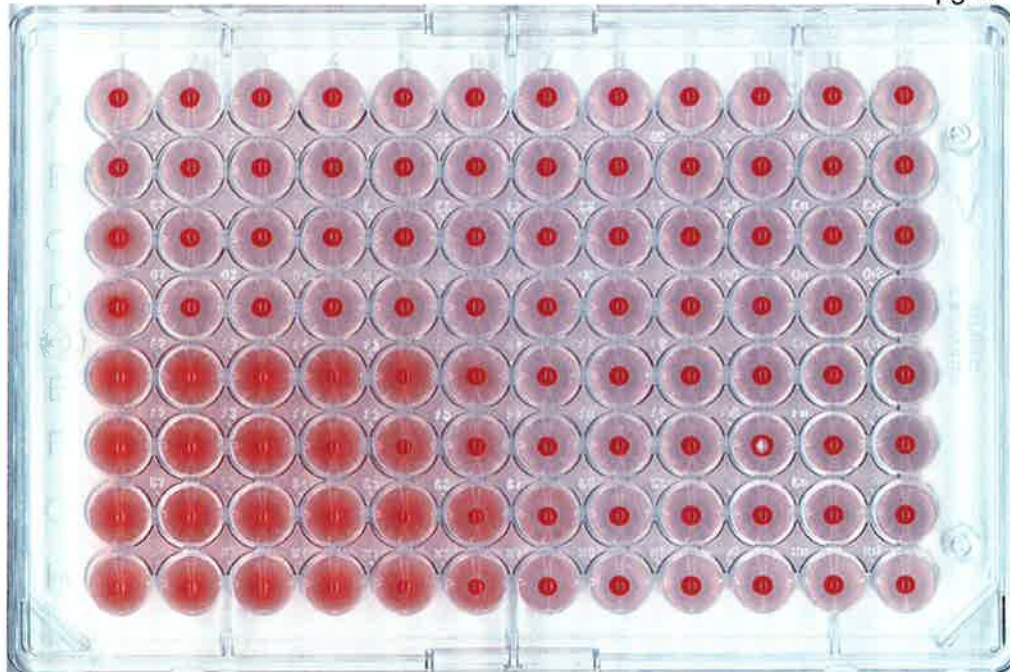
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**Haemagglutination (human blood, group 0), Lot 221501**
***Lens culinaris* agglutinin (LCA)**

Column	1	2	3	4	5	6	7	8	9	10	11	12
Conc	333	167	83	42	21	10	5.2	2.6	1.3	0.6	0.3	0.16 $\mu\text{g/mL}$



Row A,B	Blank
Row C,D	With 20 mM $\alpha$ -metyl-D-mannosid
Row E,F	With 2 mM $\alpha$ -metyl-D-mannosid
Row G,H	Only lectin without $\alpha$ -metyl-D-mannosid (agglutination control)

Plate scanned:

Date: 2016-04-20

 Signature: *Muhammad Hassan*

## IEF analysis *Lens culinaris* lectin, Lot 221501

Isoelectric focusing using Pharmacia Phastsystem (Amersham Biosciences).

### Materials

Phastgel IEF 3-9

Reference proteins pH 3.5 – 9.3 (IEF Stdrd)

Amylglucosidase -	3.50 pl
Methyl red (dye) -	3.75 pl
Trypsin inhibitor -	4.55 pl
b-Lactoglobulin A -	5.20 pl
Carbonic anhydrase B (bovine) -	5.85 pl
Carbonic anhydrase B (human) -	6.55 pl
Myoglobin, acidic band -	6.85 pl
Myoglobin, basic band -	7.35 pl
Lentil lectin, acidic -	8.15 pl
Lentil lectin, middle -	8.45 pl
Lentil lectin, basic -	8.65 pl
Trypsinogen -	9.30 pl

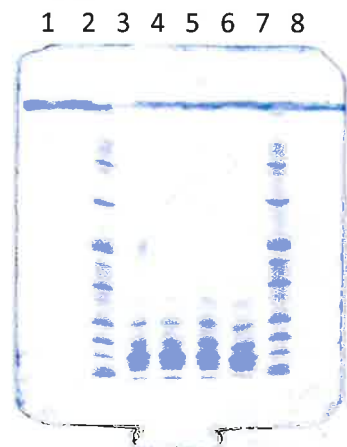
### Method

The proteins were diluted to 4 mg/ml in milliQ water supplemented with 30 % saccarose. The gels were fixed for 3 min, washed and stained with IEF staining sol. for 10 min at 50°C and then destained.

#### Program

Sample applicator down at	2.2	0Vh				
Sample applicator up at	2.3	0Vh				
SEP 2.1	2000V	2.5mA	3.5W	15°C	75Vh	
SEP 2.2	200V	2.5mA	3.5W	15°C	15Vh	
SEP 2.3	2000V	2.5mA	3.5W	15°C	410Vh	

### Result



Lane 2. IEF standard pH 3.5 – 9.3

Lane 3. Lot 221501

Lane 4. Lot 221501

Lane 5. Lot 220312 reference

Lane 6. Lot 220312 reference

Lane 7. IEF standard pH 3.5 – 9.3

Analysis performed by

*Doreen Heinrich*  
Doreen Heinrich, 2016-04-21