



**ANTI-H LECTIN**  
**ULEX EUROPAEUS LECTIN FOR SLIDE AND TUBE TESTS**

**SUMMARY**

The H antigen is a basic blood group antigen present in human beings. There is considerable variation in the H antigen content in different individuals of the same ABO group but the general pattern indicates their strength as O>A<sub>2</sub>>A<sub>1</sub>,B>B>A,>A,B. Water soluble H substance can also be demonstrated in saliva or body fluids of individuals who are secretors.

Human red blood cells that do not agglutinate with Anti-H lectin are classified as Bombay Phenotype (Oh). The Bombay Phenotype is more common in India than other parts of the world and the estimated gene frequency of Oh phenotype in Bombay is 0.0066%.

**PRESENTATION**

REF	10210005
ERYBANK® Anti-H lectin	5 ml
Pack insert	1

**REAGENT**

ERYBANK® Anti-H lectin is a ready to use purified extract of Ulex europaeus seeds. It contains a phytohaemagglutinin, which is virtually specific for the H antigen on human red blood cells.

ERYBANK® Anti-H lectin is used for recognition of the H antigen on human red blood cells. It is useful, especially for assessing the H secretor status of group 'O' individuals and also in differential grouping of A<sub>int</sub> subgroup along with Anti-A, lectin.

**REAGENT STORAGE AND STABILITY**

- Store the reagent at 2-8°C. DO NOT FREEZE.
- The shelf life of the reagent is as per the expiry date mentioned on the reagent vial label. Once opened the shelf life of the reagent vial is as described on the reagent vial label provided it is not contaminated.

**PRINCIPLE**

Human red blood cells possessing the H antigen will agglutinate in the presence of seed extract (lectins) containing phytohaemagglutinin specifically directed towards it. Water soluble H substance present in saliva neutralises the agglutination sera for H lectin. Agglutination of red blood cells / Neutralization of agglutinating sera for H lectin by saliva is a positive test result and indicates the presence of H substance on/in the red cell / saliva respectively.

No agglutination / Neutralization of the agglutinating sera for H lectin is a negative test result and indicates the absence of H substance on / in the red cell / saliva respectively.

**PRECAUTIONS**

- In vitro diagnostic reagent for laboratory and professional use only. To be used by a qualified personnel. Not for medicinal use.
- The reagent contains sodium azide 0.1% as preservative. Avoid contact with skin and mucosa. MSDS available on request.
- Extreme turbidity may indicate microbial contamination or denaturation of protein due to thermal damage. Such reagents should be discarded.
- Reagents are not from human source, hence contamination due to HBsAg, HIV and HCV is practically excluded.
- It is necessary to use the dropper provided in the reagent vial to dispense a reagent drop.
- It is advisable to wear gloves and safety spectacles and handle test specimens of human origin with caution.
- Do not use damaged or leaking reagents.
- Special protective measures, conditions for disposal and disinfection should be implemented in accordance with local regulations.

**SAMPLE COLLECTION AND PREPARATION**

**For recognition of H antigen on human red blood cells**

No special preparation of the patient is required prior to sample collection by approved techniques.

Samples should be stored at 2-8°C, if not tested immediately. Do not use haemolysed samples.

Anticoagulated blood using various anticoagulants should be tested within the below mentioned time period:

EDTA or Heparin	:	2 days
Sodium citrate or sodium oxalate	:	14 days
ACD or CPD	:	28 days

**For assessing secretor status in human saliva**

- a) Collect about 2 ml of fresh saliva in a glass tube and incubate in a boiling water bath for 10 minutes.
- b) Centrifuge at 3400 rpm (1000 g) for 10 minutes.
- c) Use the clear supernatant immediately for the study or freeze immediately if to be tested later.

**ADDITIONAL MATERIAL REQUIRED FOR SLIDE AND TUBE TESTS**

Glass slides (50 x 75 mm), Test tubes (10 x 75 mm), pipettes, isotonic saline, Centrifuge, Timer, Mixing sticks. "O" phenotype red blood cells positive for H antigen.

**PROCEDURE**

Bring all reagents and samples to room temperature before testing.

**Slide Test**

1. Place one drop of ERYBANK® Anti-H lectin on a clean glass slide.
2. Add 50µl of whole blood to be tested on the slide and mix well with a mixing stick uniformly over an area of approximately 2.5 cm<sup>2</sup>.
3. Rock the slide gently, back and forth.
4. Observe for agglutination macroscopically at two minutes.

**Tube Test**

1. Prepare a 5% suspension of the red cells to be tested in isotonic saline.
2. Place one drop of ERYBANK® Anti-H lectin into a test tube.
3. Pipette into the test tube, 50µl of the test red cell suspension and mix well.
4. Centrifuge for 1 minute at 1000 rpm (125 g) or 20 seconds at 3400 rpm (1000 g).
5. Gently resuspend the cell button, observing for agglutination macroscopically.

**Tube Test (Secretor Status)**

1. Take two clean glass test tubes labeled as No. 1 and 2.
2. Place two drops of ERYBANK® Anti-H lectin into each tube.
3. Pipette 100µl of saliva in tube No.1 and add 100µl of saline in tube No.2 and mix well.
4. Incubate at room temperature for ten minutes.
5. Add 50µl of 5% cell suspension of known "O" red cells reactive with ERYBANK® Anti-H lectin to both the tubes, mix well and Incubate at room temperature for 5 minutes.
6. Centrifuge for one minute at 1000 rpm (125 g) or 20 seconds at 3400 rpm (1000 g).
7. Gently resuspend the cell button in each test tube observing for agglutination macroscopically.

**INTERPRETATION OF RESULTS****Slide and Tube Tests**

Agglutination is a positive test result and indicates the presence of H antigen.

No agglutination is a negative test result and indicates the absence of H antigen and the red cells being of Bombay phenotype.

**Tube Test (Secretor Status)**

Agglutination of the red cells in tube No.1 indicates that the Anti-H lectin has not been neutralised and the patient is a non-secretor.

No agglutination of the red cells in tube No. 1 indicates the Anti-H lectin has been neutralised and the patient is a secretor.

The above interpretation is valid only if the tube No.2 shows agglutination.

**REMARKS**

1. Do not interpret peripheral drying or fibrin strands as agglutination.
2. It is recommended that known negative and positive cells must be included as controls with each test series.
3. As undercentrifugation or overcentrifugation could lead to erroneous results, it is recommended that each laboratory calibrate its own equipment and the time required for achieving the desired results.

**PERFORMANCE CHARACTERISTICS**

127 whole blood samples with known cell phenotype were assessed with the ERYBANK® Anti-H lectin reagent. 100% specificity of the ERYBANK® Anti-H lectin reagent versus the expected results was demonstrated with the samples used for assessment. The general pattern of reactivity of ERYBANK® Anti-H lectin with the cells of various phenotype used was observed to be O>A<sub>2</sub>>A<sub>2</sub>B>B>A<sub>1</sub>>A<sub>1</sub>B as expected.

**WARRANTY**

This product is designed to perform as described on the label and the package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

**BIBLIOGRAPHY**

1. Lee H.H., Rouger P., Germain C., Muller A. & Salmon C. (1983). , The production and standardisation of monoclonal antibodies as AB blood group typing reagents, Symposium of International Association of Biological Standardisation on monoclonal antibodies.
2. Race R. & Sanger R., Blood Groups in Man., 6th Ed., Blackwell Science, Oxford 1975.
3. Technical Manual American Association of Blood Banks, 9thEd., 1985. 127-153.
4. Data on file: Tulip Diagnostics (P) Ltd.

#### SYMBOL KEYS

	Temperature limitation		Manufacturer	<b>LOT</b>	Batch Number/Lot Number		This side up		
	Use by		Consult Instructions for use	<b>EC</b>	<b>REP</b>	Authorised Representative in the European Community	<b>REAGENT</b>	Description of reagent	
	Date of Manufacture	<b>REF</b>	Catalogue Number	<b>IVD</b>	<i>In vitro</i> Diagnostic Medical Device			<b>PS</b>	Production Site



**PS**

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**EC** **REP**

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