

**NATIONAL INSTITUTE OF BIOLOGICALS
NOIDA
BLOOD REAGENT LABORATORY**

Monographs for Blood Grouping Lectins

Anti-A₁

Source

Anti-A₁ (Lectin) is an extract of *Dolichos biflorus* seeds prepared in a saline diluent.

Of various subdivisions of A antigen (i.e., A₁, A₂, A₃, A_m, A_x, etc), A₁ and A₂ are encountered most frequently. Group A red blood cells which are agglutinated with Anti-A₁ Lectin are said to be sub group A₁ and those which are not agglutinated by Anti-A₁ Lectin fall into subgroups weaker than A₁, the majority being classified as A₂. Approximately 80% of the population of blood group A is A₁ while the remaining 20% are A₂ or weaker subgroup.

In the undiluted state, the lectin extract of *Dolichos biflorus* reacts as Anti-A, since it agglutinates both A₁ and A₂ cells. When prepared at an appropriate dilution, however, the lectin reacts directly with A₁ and A₁B but not A₂ or A₂B red cells.

Principle of the Test

Direct agglutination of group A and AB red blood cells by Anti-A₁ (Lectin) indicates the presence of the A₁ or A₁B antigens. No agglutination, indicates that the red blood cells carry a weaker form of the A antigen.

Anti-A₁ (Lectin) reagents are tested for their Physical Appearance, Color, Potency, Intensity, Specificity, Rouleaux, and Haemolysis and Prozone parameters and have values at least equal to the appropriate International Standards or suitable reference preparation used.

Physical Appearance and Color:

Anti-A₁ (Lectin) reagent should be clear with no turbidity, precipitate, particles or gel formation by visual inspection. Anti-A₁ (Lectin) blood grouping reagents are colorless liquids.

Potency

It is defined as the reciprocal of the greatest reagent dilution for which the reaction is graded at 1+. Anti-A₁ (Lectin) reagent should have an average potency value at least equal to that of the reference preparation.

Type of Reagent	Test Red Blood Cells	Titer
Anti-A ₁ (Lectin)	A ₁ (2 cells) A ₁ B (2 cells)	1: 8 to 1:16 (1+) 1: 4 to 1:8 (1+)

Intensity

It is defined by the appearance of agglutination obtained after mixing undiluted Anti-A₁ (Lectin) reagent with washed red blood cell suspension. For this 1 volume of the reagent is added to 1 volume of 40% red cell suspension on a slide and mixed for 2 minutes. At the end of 2 minutes, reaction grade is compared with the strength of reaction obtained with the reference preparation.

Type of Reagent	Test Red Blood Cells	Intensity
Anti-A ₁ (Lectin)	A ₁ (2 cells)	2+ to 3+

Specificity

Reagent is tested with red cell suspension which exhibits the antigen corresponding to the reagent antibody and yields positive reaction and red cell suspension which lacks the antigen corresponding to reagent antibody yields negative reaction.

For this 1 volume of Anti-A₁ (Lectin) reagent is mixed in a test tube with 1 volume of 2-3% red cell suspension which exhibits the antigen corresponding to the reagent antibody and red cell suspension which lacks the antigen corresponding to reagent antibody. Centrifuge the tube at 1000rpm for 1 minute. Tube with corresponding antigen gives positive reaction and the tube which lacks the antigen gives negative reaction.

Type of Reagent	Test Red Blood Cells	Specificity
Anti-A ₁ (Lectin)	A ₁ (2 cells) A ₁ B (2 cells) A ₂ (2 cells) A ₂ B (2 cells) B (2 cells) O (2 cells)	Positive Positive Negative Negative Negative Negative

Reactivity

Haemolysis

It is defined as the release of hemoglobin from the red cells as a result either of osmotic effect or of the breaking up of the red blood cells. No immune hemolysis should be observed with the red blood cells used for specificity.

Type of Reagent	Test Red Blood Cells	Haemolysis
Anti-A ₁ (Lectin)	A ₁ (2 cells) A ₁ B (2 cells) A ₂ (2 cells) A ₂ B (2 cells) B (2 cells) O (2 cells)	Absent Absent Absent Absent Absent Absent

Rouleaux

It is defined as a form of pseudo agglutination in which the red blood cells look like pile of coins. The red blood cells lacking the antigen corresponding to the reagent antibody should not show rouleaux formation. For this 1 volume of Anti-A₁ (Lectin) is mixed in a test tube with 1 volume of 2-3% red cell suspension which lacks the antigen corresponding to reagent antibody and the tube is centrifuged at 1000rpm for 1 minute and observed microscopically for rouleaux formation.

Type of Reagent	Test Red Blood Cells	Rouleaux
Anti-A ₁ (Lectin)	A ₂ (2cells)	Absent
	A ₂ B (2 cells)	Absent
	B (2 cells)	Absent
	O (2 cells)	Absent

Prozone

It is defined as a phenomenon in which negative reactions are obtained with low dilutions of an antibody, while a positive reaction is obtained with higher dilutions. For this 1 volume of Anti-A₁ (Lectin) is mixed with 1 volume of 2-3% red cell suspension in test tubes which exhibit the antigen corresponding to the reagent antibody for “15 minutes”, “30 minutes”, and “60 minutes”. If the reaction grades are the same or increase as the incubation time increases, no prozone is present and if the reaction grades decrease as the incubation time increases, prozone is present.

Type of Reagent	Test Red Blood Cells	Specificity
Anti-A ₁ (Lectin)	A ₁ (2 cells)	Absent
	A ₁ B (2 cells)	Absent

Expiration date. The expiration date for Anti-A₁ (Lectin) reagent is not less than 1 year at 2° to 8°C.

Storage. Store at a temperature between 2° to 8°C.