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Inhibition Reactions of Ten Nonspecific Seed Extracts and Their Conversion into Group Specific Lectins

Rosy Sachdeva and V. Bhalla

Department of Anthropology, Panjab University, Chandigarh 160014, India

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ABSTRACT Inhibition reactions of 10 nonspecific lectins have been examined using a panel of 22 simple and derivative sugars. The results of haemagglutination inhibition reactions suggest that all ten extracts, which apparently behave as nonspecific lectins, in fact, are highly specific with respect to the binding of saccharides. They possess multiple binding sites and could be converted into blood group specific lectins in the presence of certain inhibiting sugar (s). Six seed extracts namely *Terminalia bellirica, Terminalia citrina, Eucalyptus staigriana, Ficus infectoria, Morus alba* and *Vigna catjang* after partial inhibition with certain carbohydrates, behaved like anti-(A+B). Three lectins (*Cassia siamea, Trichosanthus anguina, Ficus infectoria*) could be converted into anti-(A+H) specific and one (*Acalypha indica*) into anti-(B+H). One lectin each could be converted into anti-A (*Ficus infectoria*) and anti-H (*Trichosanthus anguina*), While two lectins (*Eugenia jambolana, Terminalia bellirica*) could be converted into anti-B specific reagents after treatment with appropriate inhibiting sugars.

INTRODUCTION

Lectins differ widely in their specificity in cell agglutination reactions and in the susceptibility of these reactions to inhibition by saccharides. Most plant extracts agglutinate the erythrocytes of all human blood types and are usually referred to as nonspecific lectins. It has been shown that among the large group of so called nonspecific lectins, there are many which are not really nonspecific. Inhibition tests with a variety of sugars show that they are highly specific with respect to binding of saccharides. The sugar specificity of these lectins is not necessarily related to blood group determinants. In many cases, these lectins interact with erythrocytes by binding to saccharide receptors different from blood type determinants and such receptors are present on all red blood cell surfaces.

In this paper, serological specificities of ten seed extracts which yielded lectins that agglutinated human erythrocytes of all blood types have been examined using a panel of 22 sugars. The study is an attempt to explore the possibilities of converting the so called nonspecific lectins into group specific lectins by treatment with appropriate inhibiting sugars.

MATERIAL AND METHODS

The seed extracts were prepared following the standard technique (Hakim and Bhatia, 1965). Sodium azide (.01%) was added as preservative and the extracts were stored at -20° C. The agglutination reactions with saline washed human red cells were carried out at room temperature. The extracts were titrated against all ABO blood group red cells.

22 Simple and derivative sugars : D (-) fructose, α-L-Rhamnose, α-D-glucose pentaacetate, α-D-glucose, α-lactose, D(+)-cellobiose, 2-Deoxy-D-glucose, N-acetyl-D-glucosamine, Maltose, D(+) xylose, D(+) Raffinose pentahydratre, D(-) Ribose, Salicin, D(-) Arabinose, L(+) Arabinose, Dulcitol, D(+) galactose, α-D (+) Melibiose, D(+) Mannose, α-L (-) Fucose, N-acetly-D-galactosamine and Sucrose; were used to study their inhibitory effect on haemagglutinating activity of these lectins against human red cells. The sugar inhibition tests were carried out following the standard technique (Bhatia and Boyd, 1962), using 0.2 molar concentration of sugar solutions.

RESULTS AND DISCUSSION

Table 1 shows heamagglutination reactions

of ten seed extracts with adult human erythrocytes. Although all ten extracts agglutinated the human erythrocytes of all blood types, differences in their serological specificities are evident from disparities in the avidity and titre of their reactions. The results of sugar inhibition tests (Table 2) show that all ten lectins examined could be completely or partially inhibited by the addition of one or more sugars. None of the ten extracts was inhibited by α-D-glucose pentaacetate, maltose and sucrose. 19 sugars utilised in the present investigation inhibited the reactivity of one or more lectins. It was observed that Monosaccharide α-L (-) fucose, Disaccharide D-(+) Melibiose and galactose containing trisaccharide D(+) Raffinose pentahydrate inhibited the maximum number of lectins *i.e.*, 5 each whereas disaccharide cellobiose, modified glucose derivative sugar 2-Deoxy-D-glucose, Monosaccharides D(-) Ribose and D(+) Mannose inhibited the reactivity of one lectin each.

The results of haemagglutination-inhibition reactions suggest that all ten extracts, which apparently behave as nonspecific lectins, infact, are highly specific with respect to the binding of saccharides. They possess multiple binding sites and could be converted into blood group specific lectins in the presence of certain inhibiting sugar (s). Six (Table 2) seeds extracts namely *Terminalia bellirica, Terminalia citrina, Eucalyptus staigriana, Ficus infectoria, Morus alba* and *Vigna catjang* after partial inhibition with certain carbohydrates, behaved like anti-(A+B). Three lectins (*Cassia siamea, Trichosanthus anguina, Ficus infectoria*) could be converted into anti-(A+H) specific and one (*Acalypha indica*) into anti-(B+H). One lectin each could be converted into anti-A (*Ficus infectoria*) and anti-H (*Trichosanthus anguina*), while two lectins (*Eugenia jambolana, Terminalia bellirica*) could be converted into anti-B specific reagents after treatment with appropriate inhibiting sugars.

Bird (1976), on the basis of inhibition studies with saccharides, divided the anti-H type reagents into two groups:

1). Those inhibited by L (-) fucose.

2). Those not inhibited by L(-) fucose, but by various other sugars such as diacetyl-chitobiose, an N-acetyl-D-glucosamine derivative and salicin, a glucose derivative.

According to this scheme, four extracts (*Cassia siamea, Ficus infectoria, Eugenia jambolana* and *Terminalia bellirica* qualify to be classified as anti-H type lectins, in the sense, that they were inhibited by L(-) fucose and/or Salicin, the carbohydrates previously found to inhibit anti-H lectins. However, these extracts

Table 1: Heamagglutination reactions of ten seed extracts with adult human erythrocytes

S.	*Name of*	*Blood group of Red cells*				*Titre*	*Time* (in mins)*	
No.	*extract*	*O*	*A*	*B*	*AB*		*a*	*b*
1.	*Terminalia bellirica*	3+	3+	3+	3+	4	5	25
2.	*Morus alba*	2+	2+	2+	2+	4	12	25
3.	*Trichosanthus anguina*	4+	4+	4+	4+	64	3	10
4.	*Acalypha indica*	3+	3+	3+	3+	4	10	20
5.	*Eugenia jambolana*	2+	2+	2+	2+	4	10	20
6.	*Vigna catjang*	3+	3+	3+	3+	16	7	20
7.	*Terminalia citrina*	3+	3+	3+	3+	8	2	10
8.	*Eucalyptus staigriana (seed coat)*	2+	2+	2+	2+	4	4	15
9.	*Cassia siamea*	2+	2+	2+	2+	4	12	25
10.	*Ficus infectoria*	2+	2+	2+	2+	4	15	25

*Time** (a) : indicates the time when reaction was first visible to naked eye
(b) : indicates the time taken by completed reaction

Table 2: Inhibition reactions of lectins

S. No.	*Name of lectins*	*Name of sugars which completely inhibited heamagglutinating activity of lectin*	*Partial inhibition of lection*	
			Name of inhibiting sugar	*Conversion of nonspecific lectin into*
1.	*Terminalia bellirica*	α-L Rhamnose N-acetyl-D-glucosamine D(-) Ribose D(-)Arabinose Dulcitol α-L(-)fucose	D(+)xylose Salicin α-D (+) -Melibose	anti-(A+B) anti-(A+B) anti-B
2.	*Morus alba*	D(-)fructose α-D-glucose α-lactose α-D(+)-Melibiose	D(+)-Raffinose pentahydrate	anti-(A+B)
3.	*Trichosanthus anguina*	α-Lactose D(+)Raffinose pentahydrate α-D(+)-Melibiose	α-L(-)fucose N-acetyl-D glucosamine*	anti-H anti-(A+H)
4.	*Acalypha Indica*	α-D(+)-Melibiose N-acetyl-D-galactasamine	Dulcitol D(+)galactose	anti-(B+H) anti-(B+H)
5.	*Eugenia jambolana*	L(+)Arabinose α-L(-)fucose α-D-glucose	D(-)fructose α-L-Rhamnose Dulcitol	anti-B anti-B
6.	*Vigna catjang*	α-Lactose L (+) Arabinose D(+) galactose α-D-(+)Melibiose N-acetyl-D-galactosamine	D-(+)- Raffinose pentahydrate	anti-(A+B)
7.	*Terminalia citrina*	--	N-acetyl-D-galactosamine	anti-(A+B)
8.	*Eucalyptus staigriana* (seed coat)	α-D-glucose	N-acetyl-D-galactosamine	anti-(A+B)
9.	*Cassia siamea*	D(-)fructose α-D-glucose D(+)cellobiose Salicin D(+)Raffinose pentahydrate Dulcitol L(+)Arabinose α-L(-)fucose	2-Deoxy-D-glucose N-acetyl-D-glucosamine	anti-(A+H) anti-(A+H)
10.	*Ficus infectoria*	D(+)Raffinose pentahydrate Dulcitol α-L(-)fucose	α-D-glucose 2-Deoxy-D-glucose D(-)Arabinose D(+)Mannose D(-) Fructose N-Acetyl-D-glucosamine L(+)-Arabinose	anti-(A+B) anti-(A+B) anti-(A+B) anti-(A+H) anti-(A+H) anti-A anti-A

*Sugar inhibition without complete neutralization of haemagglutinating activity of lectin [Reaction was O(4+) A(4+) B(1+)]

reacted equally strongly with all ABO blood groups and therefore, cannot be used for differentitation of O and A_2 blood from the rest. Further investigations should throw more light on their potential use as tools for the study of weaker variants of blood group antigens.

REFERENCES

Bhatia, H.M. and Boyd, W.C. : Inhibition reactions of fourteen nonspecific seed extracts. *Transfusion*, **2** : 106-109 (1962).

Bird, G.W.G. : Heterogeneity of anti-H lectins. *Rev. Fr. Transfus. Immuno-hematolagie.*, **19** : 175-183 (1976).

Dunsford, I. and Bowley, D.C. : *Techniques in Blood Grouping*. Oliver and Boyd, London (1967).

Hakim. S.A. and Bhatia, H.M. : Serological specificity to anti-A on the basis of inhibition reactions. *Ind. J. Med. Res.*, **53** : 291-297 (1965).

Makela, O. : *Studies in Haemagglutinins in Leguminosae Seeds*. Academic Dissertation. Helsinki (1957).