“New” lectins for the identification of erythrocyte cryptantigens and the classification of erythrocyte polyagglutinability: *Medicago disciformis* and *Medicago turbinata*

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SUMMARY The seeds of *Medicago disciformis* and *Medicago turbinata* contain lectins for T or Th but not Tk or Tx red cell cryptantigens. Extracts of *Medicago disciformis* seeds are a useful addition to a panel of lectins used in the classification of red cell cryptantigens and of red cell polyagglutinability.

Cell membrane cryptantigens are of interest in clinical pathology because they may be exposed by the action of microbial enzymes, or by incomplete biosynthesis of membrane carbohydrates due to the development, by somatic mutation, of an abnormal clone of erythrocyte precursor cells.1

Lectins are essential for the identification of erythrocyte antigens and cryptantigens and for the classification of erythrocyte polyagglutinability.2 Use of a battery of selected lectins not only facilitates classification but also leads to the discovery of “new” forms of erythrocyte polyagglutinability.

Two very useful lectins are those of the leguminous plants *Arachis hypogaea* (peanut)3 and *Vicia cretica*.3 The *Arachis hypogaea* lectin reacts with at least four cryptantigens: T,2 Tk,4 Th5 and Tx6, exposed by various microbial enzymes, whereas the *Vicia cretica* lectin reacts with T and Th, but not Tk or Tx.3,6 We now report two “new” lectins which promise also to be of value in the classification of erythrocyte polyagglutinability. The lectins have been obtained from the seeds of *Medicago disciformis* DC and *Medicago turbinata* (L) All (syn *Medicago aculeata* Gaertn) (NO Leguminosae).

Material and methods

The seeds were obtained from the Seed Bank of the Royal Botanic Gardens, Kew. Extracts were prepared as described by Boyd and Reguera.7 Polyagglutinable red cells were obtained from a comprehensive panel held in our laboratory. Agglutination and agglutination-inhibition tests were done by both tile and tube techniques as described by Bird and Wingham.8 Simple sugars known to be components of the human red cell membrane were prepared as 2% aqueous solutions for simple monosaccharides—for example L-fucose, and as 1% aqueous solutions for aminosugars—for example, N-acetyl-D-galactosamine.

Results

The agglutination reactions of extracts of *Medicago disciformis* seeds with various red cell cryptantigens are recorded in Table 1, and compared with those of some other lectins currently applied to the classification of cryptantigens associated with red cell polyagglutinability. *Medicago turbinata* seed.

Table 1 Reactions of the *Medicago disciformis* lectin and other lectins on various erythrocyte cryptantigens

<table>
<thead>
<tr>
<th>Lectins</th>
<th>Red cells</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>T</td>
<td>Tk</td>
<td>Th</td>
<td>Tx</td>
<td>Tn</td>
<td>PT</td>
</tr>
<tr>
<td>Arachis hypogaea</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Vicia cretica</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Medicago disciformis</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Glycine soja</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>BS II</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

PT = papain-treated erythrocytes. BS II = a second isolectin from the seeds of *Bandeiraea simplicifolia*. + = agglutination. - = no agglutination.
Table 2  Titre scores of the Medicago disciformis and Medicago turbinata lectins on polyagglutinatable red cells

<table>
<thead>
<tr>
<th>Red cells</th>
<th>Medicago disciformis</th>
<th>Medicago turbinata</th>
</tr>
</thead>
<tbody>
<tr>
<td>T</td>
<td>83</td>
<td>73</td>
</tr>
<tr>
<td>Tx</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Th</td>
<td>15</td>
<td>2</td>
</tr>
<tr>
<td>Tx</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Tn</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

extracts are haemolytic in the undiluted state. This tendency can be just diluted out, leaving powerful haemagglutinating activity. Titre scores of both lectins against various polyagglutinatable red cells are recorded in Table 2. Neither lectin is inhibited by L-fucose, D-galactose, N-acetyl-D-glucosamine or N-acetyl-D-galactosamine.

Extracts of the seeds of the undermentioned Medicago species do not contain erythrocyte agglutinins: M coronata, M echinus, M intertexta var ciliaris, M littoralis, M lupulina, M marina, M minima, M murex, M orbicularis, M rugosa, and M sativa (lucerne, alfalfa).

Discussion

The Arachis hypogaea lectin, which is widely used in the classification of red cell polyagglutinability, has a "broad-spectrum" of activity and reacts with various cryptantigens exposed by microbial enzymes: T, Tk, Th and Tx. The Vicia cretica lectin\(^1\) reacts well with T or Th but not at all with Tk or Tx, and is therefore a very useful lectin for the investigation of polyagglutinatable red cells. Vicia cretica seeds are not easy to obtain. The Medicago disciformis lectin, which also agglutinates T or Th but not Tk or Tx, may be used instead. The Medicago turbinata lectin is not quite as good, because although it reacts very strongly on T-transformed red cells it is not as good as Medicago disciformis with Th. The Medicago disciformis lectin is strong and avid and is a useful addition to the panel of lectins used in the classification of cryptantigens and of red cell polyagglutinability. The Medicago lectins are not inhibited by any of the simple sugars which are components of the red cell membrane. Tests with more complex sugars will form a part of a separate investigation.

We thank the Curator, Seed Bank, Royal Botanic Gardens, Kew, for gifts of the various Medicago seeds. Dr GWG Bird is supported by a grant from the Medical Research Council.

References

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doi: 10.1136/jcp.36.2.195

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