Demonstration of Blood Group Substance H and Galactosyl Groups on Candida albicans

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(C. albicans was cultured on Sabouraud medium using a peptone prepared by autolysis from pancreas of the cow (Serva, Heidelberg, GFR); this preparation was found not to contain blood group active materials. Saline suspensions of the blastospores—before and after treatment with 0.01% pronase (Calbiochem, California, USA) for 5’ min—were mixed with equal amounts of the heterophile agglutinins on microscope slides and shaken gently for approximately 1 min. Agglutination reactions were read macroscopically and microscopically.

Results and Discussion

As will be seen from the Table, 8 out of 15 plant extracts tested were found to agglutinate the blastospores of C. albicans. Two of these extract (Arachis, Robinia) caused agglutination only if the yeast cells had previously been treated with pronase; two others (Evonymus, Concanavalin A) reacted distinctly stronger with pronase-treated cells than with nontreated cells. These findings clearly indicate that certain receptors occurring on the blastospores of C. albicans are masked by protein structure, and it may be expected that the spectrum of heterophile receptors occurring on the surface of other yeast cells can likewise be enlarged by a pretreatment with proteolytic enzymes.

Materials and Methods

C. albicans was cultured on Sabouraud medium using a peptone prepared by autolysis from pancreas of the cow (Serva, Heidelberg, GFR); this preparation was found not to contain blood group active materials. Saline suspensions of the blastospores—before and after treatment with 0.01% pronase (Calbiochem, California, USA) for 5’ min—were mixed with equal amounts of the heterophile agglutinins on microscope slides and shaken gently for approximately 1 min. Agglutination reactions were read macroscopically and microscopically.

Candida albicans, Blood groups substance H, Galactosyl groups, Heterophile agglutinins, Glycoproteins

Heterophile agglutinins prepared from plants and animals (snails, fish eggs, crabs) have been shown to react specifically with certain carbohydrate structures occurring on the surface of various cell types 1-4. These agglutinating reagents have been successfully used especially for the characterization of blood group active glycoproteins and glycolipids.

Since similar structures have already been detected on the surface of bacteria 5, it seemed reasonable to extend these studies on fungi as well.

Therefore, a pathogenic strain of Candida albicans isolated from skin lesions of a diabetic person was studied with extracts from numerous plants (see Table I) and other sources.

Table. Agglutination of Candida albicans by heterophile agglutinins of plant origin.

<table>
<thead>
<tr>
<th>No.</th>
<th>origin</th>
<th>Specificities</th>
<th>chemical</th>
<th>ref.</th>
<th>Candida albicans not treated</th>
<th>Candida albicans treated with pronase</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Soja hispida</td>
<td>incomplete pan-agglutinins</td>
<td>β-GalNAc → ?</td>
<td>7</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>Phaseolus lunatus</td>
<td>anti-A ( + C)</td>
<td>α-GalNAc →</td>
<td>8, 9</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>Ricinus communis</td>
<td>panagglutinin</td>
<td>β-galactosyl</td>
<td>3, 10, 11</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>4</td>
<td>Arachis hypogea</td>
<td>anti-T</td>
<td>β-galactosyl</td>
<td>4, 12</td>
<td>0</td>
<td>+ +</td>
</tr>
<tr>
<td>5</td>
<td>Vicia graminea</td>
<td>anti-X</td>
<td>β-galactosyl-(1,3)-GalNAc-→ 7</td>
<td>+</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>PHA (Difo)</td>
<td>anti-PHA-receptor</td>
<td>β-galactosyl ( ?)</td>
<td>13</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>waxbean</td>
<td>anti-β-galactosyl-α-glucosyl-α-mannosyl, α-arabinosyl</td>
<td>20, 21</td>
<td>++ ++</td>
<td>+ + + +</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>Robinia pseudacacia</td>
<td>panagglutinin</td>
<td>β-galactosyl ( ?)</td>
<td>3, 14, 15</td>
<td>0</td>
<td>+++</td>
</tr>
<tr>
<td>9</td>
<td>Evonymus europaeus</td>
<td>anti-B, anti-H</td>
<td>β-galactosyl-α-glucosyl-α-mannosyl, α-arabinosyl</td>
<td>2, 3, 4, 16</td>
<td>+ + + +</td>
<td>+ + + +</td>
</tr>
</tbody>
</table>

Requests for reprints should be sent to Dr. W. P. Herrmann, Universität-Hautklinik Köln, D-5000 Köln 41, Joseph-Stelzmann-Str. 9. * with technical assistance of I. WEYLAND and D. NUBER. ** Supported by the Deutsche Forschungsgemeinschaft.
Six of the 8 plant extracts agglutinating \textit{C. albicans} (no. 3—5, 7, 8, 10) are known to react specifically with $\beta$-galactosyl groups that are widely distributed non-reducing terminal components of glycoproteins, glycolipids, and polysaccharides including lipopolysaccharides. It should be noticed, however, that not all galactosyl-specific extracts tested were able to agglutinate \textit{C. albicans} (see no. 6), and that their serological specificities (as far as it concerns human red blood cells) are different. This phenomenon is most probably due to the fact that end-standing galactosyl groups occur in connection with carbohydrate structures of various compositions, and that the specificities of the extracts used here are not in all cases confined to the terminal galactosyl group alone. So far, our findings strongly suggest that several carbohydrate structures with end-standing non-reducing $\beta$-galactosyl groups are present on the surface of \textit{C. albicans}.

Concanavalin A which is known to react with various other sugar units including $\alpha$-glucosyl- and $\alpha$-mannosyl groups was also found to agglutinate \textit{C. albicans}; the specificity of this reaction remains to be clarified. Further extracts specific for N-acetyl-galactosamine groups (\textit{Helix pomatia} etc.), N-acetylglucosamine groups, $\alpha$-galactosyl groups, and neuraminic acid (not listed in the Table) did not agglutinate \textit{C. albicans}.

Among 4 extracts with anti-H specificity (no. 9—12) two were found to react with \textit{C. albicans}. These are no. 9 (\textit{Evonymus europaeus}) which is known to react with B-type rbc as well, and no. 10 (\textit{Ulex europaeus}). Absorption experiments revealed that the “anti-candida-agglutinin” of the \textit{Ulex} extract could be completely removed by absorption with O-type rbc, whereas the reactivity of the \textit{Evonymus} extract was only moderately, but distinctly reduced; absorption with B-Type rbc, however, had no effect.

These findings indicate that

a) the anti-candida-agglutinin of the \textit{Ulex} extract is serologically identical with its anti-H factor;

b) the extract from \textit{Evonymus europaeus} contained two agglutinins reacting with \textit{C. albicans} one of which was found to be serologically identical with its anti-H factor.

It may be concluded, therefore, that certain (but not all!) anti-H factors of plant origin are able to agglutinate the blastospores of \textit{C. albicans}. Consequently, this pathogenic yeast must bear receptors with a chemical structure similar, if not identical, to that of blood group substance H.

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