Association of Recurrent Vaginal Candidiasis and Secretory ABO and Lewis Phenotype

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The relationship between ABO-Le secretor phenotype and susceptibility to recurrent idiopathic vulvovaginal candidiasis (RVVC) was investigated. ABO and Lewis blood typing was done for 38 women with RVVC (case-patients) and for women in 2 control groups, consisting of 58 healthy women, who were friends identified by case-patients, and 38 race-matched, healthy hospital employees. The 3 groups were similar with regard to age and race. There was no difference in the distribution of ABO phenotype between case-patients and controls. Case-patients were more likely than members of either control group to have Le(a−b−) (nonsecretor) rather than Le(a−b+) (secretor) blood type. With combined nonsecretor Le(a+b−) phenotype and absence of the Lewis gene Le(a+b−), the relative risk of chronic recurring vulvovaginal candidiasis was 2.41–4.39, depending on the analysis technique and control group. In conclusion, there is an increased frequency of ABO-Le nonsecretor status among women with RVVC.

The etiology of recurrent vulvovaginal candidiasis remains unknown. Only rarely are secondary precipitating or underlying contributory factors identified [1].

The first step in vaginal mucosal colonization by Candida species is adherence to the vaginal epithelial cells [2]. Fucose, which inhibits in vitro binding of yeast to human vaginal epithelial cells [3–5], is also the immunodominant sugar of the H antigen of blood group O and a terminal component of Lewis antigens. H antigen is present in body fluids of secretors of the ABO blood groups, and in 94% of the secretors, there will also be Le(a) and Le(b) antigen. Although vaginal epithelium from women with recurrent candidal vaginitis have not shown increased cell affinity for adherence of Candida species in vitro [6], we hypothesized that secretor status could influence susceptibility to vaginal colonization by Candida fungi. Buford-Mason et al. [7] determined that nonsecretor status was similarly a risk factor for oral carriage of Candida albicans, and Hilton et al. [8] recently reported finding a possible genetic predisposition to recurrent candidal vulvovaginitis. Women prone to infection had a higher frequency of Le(a−b−) nonsecretor phenotype than did healthy controls. The aim of this study was to confirm the relationship between ABO-Le secretor phenotype and susceptibility to recurrent idiopathic vulvovaginal candidiasis.

Materials and Methods

Study participants were selected from patients at the Wayne State Vaginitis Clinic. They were limited to human immunodef-
ciency virus–seronegative women without diabetes mellitus or immunosuppression, who were not receiving treatment with corticosteroids.

**Case definition.** Vulvovaginal candidiasis (VVC) was defined as an episode of acute onset of vaginal soreness, irritation, vulvar burning, itching, dyspareunia, vaginal pH between 4 and 4.5, positive saline or 10% KOH wet-mount microscopic examinations, and a positive yeast culture. Women were considered to have recurrent idiopathic vaginitis if they had a documented culture-confirmed episode of acute VVC, history of more than three confirmed episodes of VVC during the year prior to being placed on antifungal maintenance therapy at the clinic, and development of a symptomatic culture-proven episode of VVC while on maintenance antifungal therapy (fluconazole, 100 mg/week; clotrimazole, 500 mg/week) or a relapse of symptomatic and culture-proven VVC within 3 months or two relapses in 6 months after cessation of 6 months of maintenance antifungal therapy. Only patients with infections caused by *C. albicans* were eligible. Patients presenting with mixed infections, including bacterial vaginosis and trichomoniasis, were excluded.

**Patient population.** Thirty-eight women meeting the case definition were identified (case-patients). Because the clinic is a tertiary clinic, it is difficult to determine the underlying population for study. Therefore, we established 2 control groups: One consisted of friends of the case-patients; the other consisted of hospital employees. It was assumed that friends would be similar with respect to sociodemographic status and behavior, including use of medical services, and would therefore reflect the underlying study population. Each case-patient was asked to identify 3 female friends for participation in the study. Friends meeting study criteria were interviewed by telephone; blood samples were obtained from the women either at the clinic or in their homes. Hospital employees provide a convenience sample of healthy women for comparison and have been used in similar studies. Female hospital employees of the same age and race as case-patients were asked to volunteer; blood samples were collected from those meeting eligibility criteria.

**Methods.** An antecubital vein blood sample was obtained for ABO and Lewis blood group phenotyping from each study participant. A clotted specimen of whole blood (red tube) was properly labeled and centrifuged for 2 min at high speed. The serum was removed and placed in a second labeled tube. ABO phenotyping (forward and reverse typing) was done by macroscopic hemagglutination: one drop of anti-A, anti-B, and anti-A, B, (Immucor, Norcross, GA) was added to a 4% test cell suspension, which was then centrifuged for 20 s at high speed.

Le(a) and Le(b) phenotyping were done by use of anti-Le(a) and anti-Le(b) murine monoclonal antibodies (Immucor). One drop of 5% cell suspension of test cells was added to a test tube filled with saline for washing of the blood cells, and the tube was centrifuged at high speed for 20 s. The saline was decanted, and one drop of anti-Le(a) added to the test tube. After 5 min of incubation at room temperature and further centrifugation at high speed for 25 seconds, macroscopic agglutination was determined. A similar procedure was done in another test tube with a suspension of 5% cell suspension and a drop of anti-Le(b). The positive and negative controls for phenotyping consisted of cells positive for or lacking the antigen (Gamma Biologicals, Houston), respectively.

**Table 1.** Distribution of blood types among women with chronic recurring vulvovaginal candidiasis (case-patients), friend controls, and race-matched hospital employees.

<table>
<thead>
<tr>
<th>Blood phenotype</th>
<th>Case-patients</th>
<th>Friends</th>
<th>Hospital employees</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>12 (32)</td>
<td>24 (41)</td>
<td>14 (37)</td>
</tr>
<tr>
<td>B</td>
<td>6 (16)</td>
<td>9 (16)</td>
<td>4 (11)</td>
</tr>
<tr>
<td>O</td>
<td>18 (47)</td>
<td>21 (36)</td>
<td>16 (42)</td>
</tr>
<tr>
<td>AB</td>
<td>2 (5)</td>
<td>4 (7)</td>
<td>4 (11)</td>
</tr>
<tr>
<td>Le(a+b−)</td>
<td>13 (34)</td>
<td>8 (14)</td>
<td>6 (16)</td>
</tr>
<tr>
<td>Le(a−b+)</td>
<td>18 (47)</td>
<td>44 (76)</td>
<td>26 (68)</td>
</tr>
<tr>
<td>Le(a−b−)</td>
<td>7 (18)</td>
<td>6 (10)</td>
<td>6 (16)</td>
</tr>
<tr>
<td>Total</td>
<td>38 (100)</td>
<td>58 (100)</td>
<td>38 (100)</td>
</tr>
</tbody>
</table>

NOTE. Data are no. (%) of subjects. Distribution of Lewis blood group was significantly different between cases and friend controls (*P* = .015).

**Data analysis.** Data were entered using Microsoft Excel [9]. SAS software (SAS Institute, Cary, NC) was used for data management and analysis [10]. Exact 95% confidence intervals were calculated around crude odds ratios, using Epi Info (version 6.0; CDC, Atlanta) [11]. For the matched-pair analysis, we calculated the Mantel-Haenszel summary odds ratio, test-based confidence intervals, and the Logit estimator with precision-based confidences intervals, using SAS software [10].

**Results**

Most case-patients (95%), friend controls (98%), and hospital employee controls (95%) were white. Cases were slightly older (mean age, 39 ± 10 years) than either friend controls (mean, 38.7 ± 8.7) or hospital employees (mean, 37.6 ± 8.1), but the differences were not statistically significant.

There was no difference in the distribution of ABO phenotype between case-patients and either control group (table 1). However, the distribution of Lewis phenotype was statistically different between case-patients and friend controls, with case-patients being more likely to have Le(a+b−) (nonsecretor) than Le(a−b+) (secretor) blood group (*χ*², *P* = .015). The distribution of Lewis phenotype among hospital employees was similar to that for friend controls; however, it was not statistically significantly different from that for case-patients (*P* = .12), probably because there were only 38 hospital employees, compared with 58 friend controls.

Combining nonsecretor phenotype with absence of the Lewis gene, we estimated risk of chronic recurring vulvovaginal candidiasis for nonsecretors compared with secretors. The risk of recurring vulvovaginal candidiasis for nonsecretors was estimated to range from 2.41 to 4.39, depending on the analysis technique and control group (table 2).

**Discussion**

In our study, women with recurrent VVC were more likely than friend controls or hospital employee controls to have the
nonsecretor phenotype, Le(a+b−) or Le(a−b−). In both control groups, secretor phenotype, Le(a−b+) predominated. Our results are similar to those of Hilton et al. [8]; however, they found a significantly higher frequency of Le(a−b−) phenotypes among women with candidal vulvovaginitis than among hospital employee controls. Our results were not significantly higher, but there were no significant differences between the distribution of Lewis phenotype between our case-patient group and that of Hilton et al. ($\chi^2$, $P = .29$), suggesting that the differences may be due to chance. Alternatively, the difference in case definitions used by or sampling discrepancies between the studies might be responsible for the variant conclusion.

To reduce ascertainment bias, we used an extremely strict definition of recurrent idiopathic vulvovaginal candidiasis and selected 2 control groups for comparison. Because case-patients were selected from women seen at a tertiary referral clinic, it was also difficult to determine the underlying population from which they came; thus, we used friends of case-patients as controls. When studying behavioral and sociodemographic characteristics, it is possible that friend controls may overmatch, thus diminishing associations between disease and outcome [12]. However, it is unlikely that case-patients would select friends on the basis of secretor phenotype, except as how this relates to racial group. While a matched analysis resulted in a slightly higher estimate of the odds ratio than an unmatched analysis, the magnitude and direction of the two estimates were very similar, and the confidence intervals overlapped. The number of participants in the second control group, hospital employees, was smaller; while this negatively affected the precision of the estimate, the odds ratio was again somewhat similar in magnitude and direction to that observed with friend controls.

Our data suggest that Lewis blood group phenotyping may be a marker of patients at risk for recurrent vulvovaginal candidiasis. If the relative risk for recurrent vulvovaginal candidiasis among nonsecretors is 3, this implies that 67% of women with recurrent vulvovaginal candidiasis and nonsecretor phenotype can attribute their vulvovaginal candidiasis to their nonsecretor phenotype. Assuming that 32% of the population has a Lewis nonsecretor phenotype, this phenotype may explain as much as 21% of chronic recurring vulvovaginal candidiasis (67% $\times$ 32%). Further studies should examine whether mucosal surface differences as well as those in vaginal secretions explain the susceptibility of some women to repeated bouts of vaginal candidiasis in the absence of other known predisposing factors.

### References