Genitourin Med 1994;70:268–272

Clinical and microbiological characteristics of symptomatic vulvovaginal candidiasis in HIV-seropositive women

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Abstract

***Objectives*—To evaluate the clinical and microbiological characteristics of symptomatic vaginal candidiasis in Human Immunodeficiency Virus (HIV)-seropositive women attending a gynaecologic out-patient clinic for sexually transmitted diseases (STDs).**

***Design*—Vaginal, rectal and oral specimens from cases and controls were cultured for *Candida spp*.**

***Subjects*—Eighty-four consecutive HIV-seropositive and 384 HIV-seronegative women with clinical signs of vulvovaginitis.**

***Setting*—A gynaecological out-patient clinic in Pavia, Italy.**

***Results*—The overall prevalence of vaginal candidiasis was 61·9% (52/84) in the cases and 32·3% (124/384; p < ·001) in the controls. After adjustment by logistic regression analysis for confounding factors (age at first intercourse, lifetime sex partners, new partner/s in the last 6 months, type of contraceptive used), HIV-seropositive patients were at higher risk for both *Candida albicans* (odds ratio = 2·5; 95% confidence interval 1·31–4·69; p = 0·006) and *Torulopsis glabrata* vaginitis (OR = 3·5; 95% CI = 1·05–11·60; p = 0·04) than controls. HIV-seropositive subjects had also increased rates of oral and rectal colonisation with *Candida spp*. Finally, the time to recurrence of vaginal infection was significantly shorter in HIV-seropositive patients than controls and was correlated with the severity of HIV-induced immunodepression.**

***Conclusions*—Vulvovaginal candidiasis is very common in HIV-seropositive women and its prevalence is correlated with the immunological status of the host. These patients have higher frequencies of *Torulopsis glabrata* vaginal infection and are more prone to recurrence than HIV-seronegative controls.**

(*Genitourin Med* 1994;70:268–272)

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Accepted for publication 10 March 1994

Introduction

It is well known that cell-mediated immunity depression correlates well with the occurrence of local or systemic candida infection.[1] In immunocompromised patients, candida organisms act as opportunistic pathogens causing clinical syndromes ranging from oral or vaginal thrush to debilitating chronic mucocutaneous candidiasis or esophagitis.[2,3] Infection with Human Immunodeficiency Virus (HIV) is probably the most common form of acquired immunodeficiency and mucosal candida infections are very common among HIV seropositive patients.[2,4,5] In particular, oral and vaginal candidiasis are considered both as indicators of the severity of the immunodeficiency and predictors of risk for serious opportunistic infections.[4–6] However, precise data on the clinical and microbiological picture and the natural history of candida vulvovaginitis among HIV seropositive women are still lacking. The purpose of this study was to evaluate the clinical and microbiological characteristics of symptomatic vaginal candidiasis among HIV seropositive women attending a centre for sexually transmitted diseases (STDs) during a two-year period.

Materials and methods

There were 84 known HIV-seropositive women with symptoms of vulvovaginal infection (vaginal pruritus, vaginal discharge, vulvovaginal irritation) attending the gynecological clinic for STDs of the University of Pavia during a two-year period (1990–1992). Of these patients, 48 (57·1%) had asymptomatic HIV infection (Category A of Centers for Diseases Control [CDC] classification),[7] 28 (33·3%) had symptomatic infection (Category B of CDC classification) and 8 (9·5%) had full-blown AIDS.

During the same period, a total of 1143 women were seen at the above institution. Of these, we considered eligible as controls 384 patients with symptoms of vulvovaginal infection and proof of negative HIV serology (HIV-serology test performed within the last two years before the study). In both cases and controls, after a detailed interview, a gynecological examination and a Pap smear were performed. Specimens collected from the vagina, rectum and the oral cavity were cultured for the diagnosis of candida infection. The specimens were collected with sterile cotton swabs and inoculated onto Sabouraud's dextrose agar plates containing gentamicin (40 μg/ml). The cultures were incubated at 30° C for 48–72 hours and then examined for yeast growth. The isolated strains were identified with either the germ tube test or auxonogram (API 20C Aux, API System, Montalieu-Vercieu, France). The susceptibility of the isolates to the most commonly used antimycotic drugs was tested using the modified Kirby-Bauer method.[8] The medium used

for the sensitivity test was the Yeast Morphology Agar (DIFCO Laboratories, Detroit, MI) and a commercially available kit (Neo-Sensitabs, A/S Rosco, Taastrup, Denmark) was used to test susceptibility to nystatin, clotrimazole, miconazole, econazole, 5-fluorocytosine and ketoconazole. The results were read after 24-hour incubation by measuring the diameter of the inhibition zone according to the manufacturers' recommendations.

All the patients with vaginal infection were treated with a 14-day course of standard topical antimycotic therapy. When given therapy, the patients were also instructed to report immediately to our Department in case of symptom relapse. In the absence of relapse, clinical and microbiological follow-up visits were carried out at four-month intervals.

In HIV seropositive patients a blood sample was also taken for white blood cell count, total lymphocyte, CD4+ and CD8+ cell counts.

Table 1 Sociodemographic and anamnestic variables in the HIV seropositive group and in controls

	HIV-positive (n = 84) N (%)	*HIV-negative (n = 384) N (%)*
Marital status		
Married	41 (48·8)	268 (69·8)
Age at first intercourse		
< 15 years	12 (14·3)	20 (5·2)
16–18 years	68 (80·9)	161 (41·9)
> 18 years	4 (4·8)	203 (52·9)‡
Previous pregnancies		
Previous term pregnancies	21 (25·0)	193 (50·3)†
Previous spontaneous abortion	8 (9·5)	40 (10·4)
Previous induced abortion	41 (48·8)	21 (5·5)†
No of lifetime sex partners		
0–1	0	153 (39·8)
2–5	23 (27·4)	199 (51·8)
> 5	61 (72·6)	32 (8·3)‡
Intravenous drug user (IVDU) (previous or current)	71 (84·5)	4 (1·0)†
Partner of IVDU	64 (76·2)	5 (1·3)†
New partner/s in the last six months	34 (40·5)	46 (12·0)†
Menstrual history		
Amenorrhoea	16 (19·0)	4 (1·0)†
Metrorrhagia	10 (11·9)	0†
Chronic pelvic pain	15 (17·9)	12 (3·1)†
Current contraceptive used		
None or "natural"	18 (21·4)	214 (55·7)†
Condom	51 (60·7)	59 (15·4)†
IUD	3 (2·6)	18 (4·7)
Pill	12 (10·4)	93 (24·2)*
Past (last year) cervicovaginal infections		
Candida	49 (58·3)	32 (8·3)†
Trichomonas	6 (7·1)	4 (1·0)*
Herpes genitalis	2 (2·4)	0*
Gonorrhoea	1 (1·2)	0

*p < 0·05; †p< 0·01; in comparison with the HIV positive group. ‡p < 0·001 by chi square for trend.
Cumulative percentages do not add up to 100 because of rounding errors.

Table 2 Frequencies of isolates in the HIV seropositive group and in controls

	HIV positive (n = 84) N (%)	*HIV negative (n = 384) N (%)*	*p*
Candida Albicans isolates			
Vaginal	38 (45·2)	106 (27·6)	0·006
Rectal	12 (14·3)	16 (4·2)	0·0004
Oral	18 (21·4)	19 (4·9)	<0·0001
Torulopsis glabrata isolates			
Vaginal	8 (9·5)	10 (2·6)	0·007
Rectal	7 (8·3)	4 (1·0)	0·0003
Oral	3 (3·6)	—	0·003
Other candida species isolates (C Tropicalis, C Parapsilosis, C Pseudotropicalis, S Cerevisiae)			
Vaginal	6 (7·1)	8 (2·1)	0·035
Rectal	2 (2·4)	2 (0·5)	0·3
Oral	1 (1·2)	—	0·4

Statistical analysis was carried out with chi square test with Yates' correction where appropriate, or chi square for trend for comparison of categorical variables. The Mann-Whitney test was used to compare continuous variables. To estimate the risk of candida infection, crude odds ratios for prevalence data and 95% confidence intervals were computed.[9] Unconditional logistic regression analysis was used to compute multivariate odds ratios adjusting for confounders.

Finally, the risk for recurrence of vaginal candidiasis was evaluated by Kaplan-Meier product limit estimates analysis and the log-rank test.

Results

The sociodemographic characteristics of the study group are reported in table 1. The mean age was 24·3 (SD 3·8) years in the cases and 25·2 (SD 3·1) in the controls (p = 0·31, Mann-Whitney test). As expected, there were some differences in sexual lifestyle between cases and controls. The majority of HIV-positive women reported intravenous drug abuse as the main risk factor for HIV infection. In HIV-negative controls, the main reasons for testing were a previous pregnancy (165/384), blood donation (65/384) or periodic screening (eg, nurses, physicians) (76/384). Miscellaneous reasons (fear of HIV infection, enrollment in screening programs, etc) were reported by the remaining patients. Twenty-five (29·7%) cases and 30 (7·8%) controls (p < 0·0001) had at least three documented episodes of candida vaginitis in the previous year.

Table 2 shows the results of the cultures for identification of *Candida spp*. Globally, vaginal cultures were positive for candida in 52 cases (61·9%) and 124 controls (32·3%) (crude odds ratio = 3·4; 95% confidence interval 2·1–5·5; p < 0·0001). All the patients found positive for candida infection had definite vulvovaginal symptomatology (vaginal discharge, pruritus, vulvar erythema, dyspareunia) and there were no evident differences in the clinical picture between cases and controls. After controlling for the confounding effect of age at first intercourse, lifetime sex partners, new partner/s in the last six months and contraceptive used, the adjusted odds ratio of *C albicans* vaginal infection in HIV-seropositive patients in comparison with controls was 2·48 (95% confidence interval 1·31–4·69; p = 0·006). The prevalence of vaginal infection was 54·2% (26/48) in patients with asymptomatic HIV infection, 71·4% (20/28) in symptomatic HIV infection and 75% (6/8) in patients with full-blown AIDS. This trend, though suggestive of increasing risk with immunodepression, was not significant (Chi square for trend = 2·60, p = 0·11).

The crude risk of *Torulopsis glabrata* vaginitis was higher among HIV-positive patients than in controls (8/84) vs 10/384; Odds ratio = 3·9; 95% confidence interval 1·5–10·3). This excess risk persisted after adjustment for con-

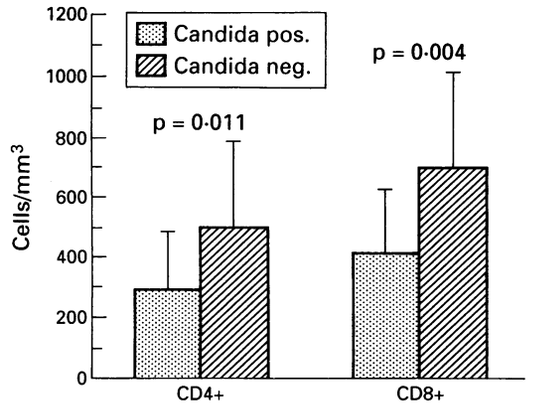

Figure 1 CD4 + and CD8 + cell counts (mean + SD) in HIV seropositive patients with and without vaginal candidiasis.

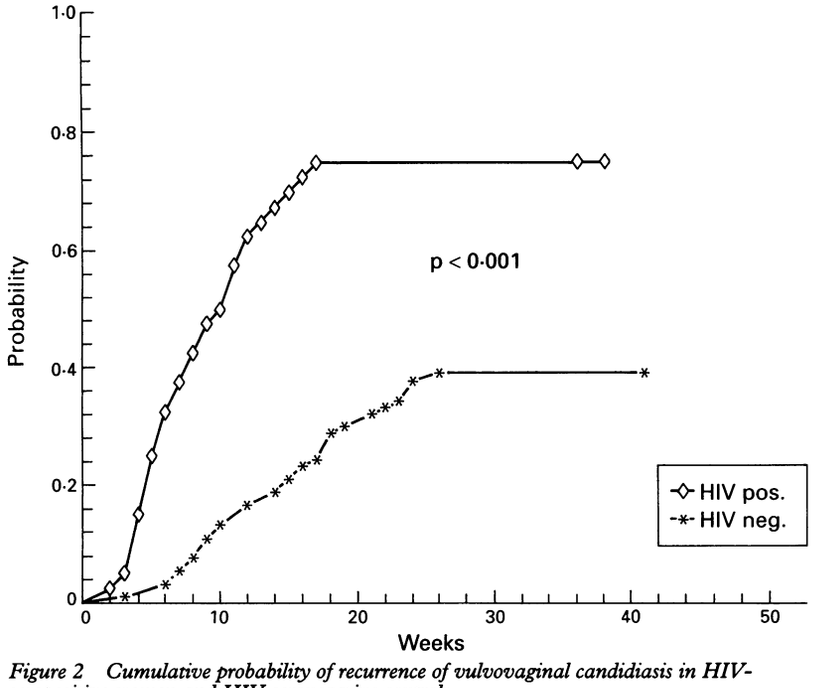

Figure 2 Cumulative probability of recurrence of vulvovaginal candidiasis in HIV-seropositive women and HIV-seronegative controls.

Table 3 Sociodemographic and anamnestic variables according to the presence of HIV-related symptoms

	*Asymptomatic** (*n* = 22) *N (%)*	*Symptomatic** (*n* = 18) *N (%)*
Marital status		
Married	12 (54·5)	10 (55·5)
Age at first intercourse		
< 15 years	4 (18·2)	3 (16·7)
16–18 years	17 (77·3)	15 (83·3)
> 18 years	1 (4·5)	0
Previous pregnancies		
0–1	12 (54·5)	10 (55·5)
⩾ 2	10 (45·4)	8 (44·4)
Previous induced abortion	16 (72·7)	12 (66·7)
No of lifetime sex partners		
0–1	0	0
2–5	11 (50·0)	8 (44·4)
> 5	11 (50·0)	10 (55·5)
Intravenous drug user (IVDU) (previous or current)	20 (90·9)	18 (100·0)
Partner of IVDU	17 (77·3)	15 (83·3)
New partner/s in the last six months	9 (40·9)	10 (55·5)
Current contraceptive used		
None or "natural"	6 (27·3)	4 (22·2)
Condom	14 (63·6)	12 (66·7)
IUD	0	0
Pill	2 (9·1)	2 (11·1)
Past (last year) cervicovaginal infections		
Candida	8 (36·4)	12 (66·7)
Trichomonas	1 (4·5)	0
Herpes genitalis	0	2 (11·1)
Gonorrhoea	0	0

*Asymptomatic and symptomatic HIV infection correspond to categories A and B + C of CDC classification, respectively. Cumulative percentages do not add up to 100 because of rounding errors. No significant differences between the two groups at the $p < 0·05$ level.

founders by logistic regression analysis (adjusted odds ratio = 3·5; 95% confidence interval 1·05–11·60; p = 0·04). Most of the patients (6 cases and 6 controls) with *Torulopsis glabrata* vaginitis reported recurrent vulvovaginal candidiasis with repeated treatment in the year before examination. Finally, vaginal specimens of HIV-positive women were found positive for *Candida spp* other than *Candida albicans* more frequently than controls (crude odds ratio = 3·6; 95% confidence interval 1·2–10·7).

Among patients with vaginal cultures positive for *Candida spp*, the frequencies of rectal and oral colonisation were 34·6% (18/52) and 32·7% (17/52) in HIV-positive women, and 16·1% (20/124; p = 0·012) and 12·1% (15/124; p = 0·0025) in controls, respectively. In few patients (6 cases and 6 controls) either oral or rectal specimens were positive for *Candida spp* whereas vaginal cultures were negative. Among cases, oral candidal colonisation was associated with clinically evident thrush in eight patients, whereas it was asymptomatic in all the control patients.

Overall, the odds ratios of rectal or oral candida colonisation were 4·6 (95% confidence interval 2·4–8·8) and 6·8 (95% confidence interval 3·5–13·3), respectively.

Regarding the results of susceptibility testing, all but two *Candida albicans* isolates whose sensitivity to 5-fluorocytosine was intermediate, were susceptible to all the antimycotic drugs tested. However, two vaginal isolates of *Torulopsis glabrata* (both from HIV-positive women) were resistant to the imidazole agents tested (clotrimazole, miconazole, econazole, ketoconazole). These two patients had previously received long courses of imidazoles (fluconazole and itraconazole) because of candida esophagitis.

Among HIV seropositive women, the immunological status as evaluated by CD4+ and CD8+ cell counts correlated well with the occurrence of vaginal candida infection (fig 1). In fact, the mean CD4+ and CD8+ counts were significantly higher in HIV positive patients with vaginal candida infection than in those with negative vaginal cultures.

We obtained information at an at least six-month follow-up in 76·9% (40/52) of the HIV-positive cases and 72·6% (90/124) of the controls. The time to clinical and microbiologically proven recurrence of vaginal candidiasis was analysed by the Kaplan-Meier product limits estimates (fig 2). The mean time to recurrence based on the date of last observation (visit or recurrence) was 15·7 (SD 2·1) weeks in the cases and 30·7 (SD 1·4) weeks in the controls ($p < 0·0001$, log-rank test). Among the 40 HIV-seropositive women followed up, 18 had symptomatic HIV infection (Categories B and C of the CDC classification system) whereas 22 were asymptomatic (Category A). The anamnestic findings were similar in these two groups (table 3). The probability of recurrence plotted against the severity of HIV infection is reported in fig 3. The mean time to recurrence was 10·1 (SD 1·94) weeks in HIV-seropositive patients with

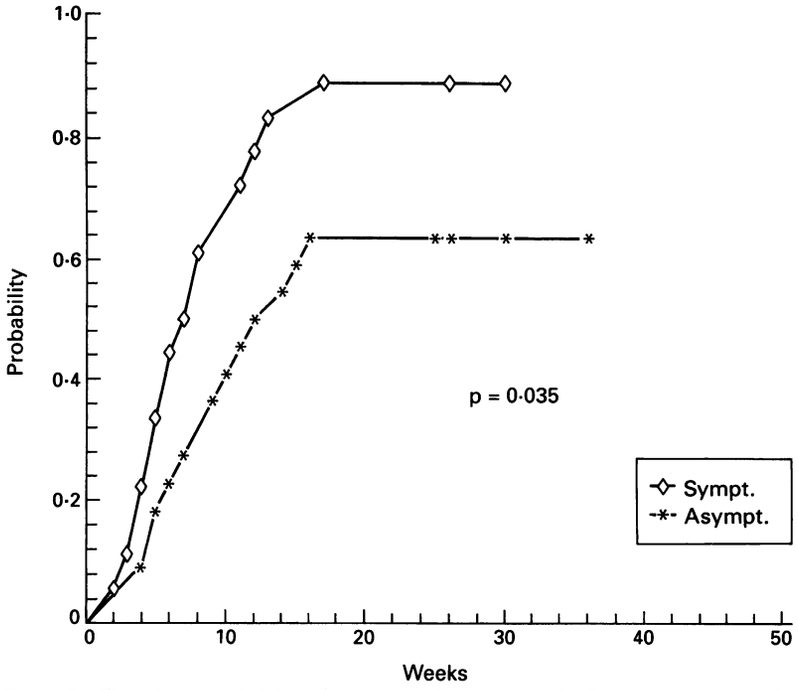

Figure 3 Cumulative probability of recurrence of vulvovaginal infection in patients with symptomatic and asymptomatic HIV infection.

symptomatic HIV infection and 19·6 (SD 3·1) in those asymptomatic with respect to HIV infection (p = 0·034, log-rank test).

Discussion

Depressed T-cell function due to drugs, immunological or systemic disease is a well recognised factor predisposing to either local or diffuse mucosal candidiasis.[1] The results of the present study clearly indicate that HIV-related immunosuppression has a considerable impact on the clinical and microbiological characteristics and the natural history of vulvovaginal candidiasis. Rhoads *et al*[4] have shown that among women with HIV infection chronic vaginal and oral candidiasis is very common and well correlated both with the severity of HIV-related immunodepression and the subsequent development of opportunisitc infections. Imam *et al*[6] have found a hierarchical pattern of mucosal candida infections among HIV-seropositive women. According to their report, the progressive colonisation of the vagina, oropharynx and the oesophagus correlates with the degree of immunodeficiency. Our series confirms that vulvovaginal candidiasis is very common in HIV-seropositive women and its occurrence correlates well with the immunological status of the host. In addition, in HIV-positive women vaginal infection has a strong tendency to recur after appropriate treatment.

Our study also shows that in HIV-positive women with symptomatic candidal vaginitis, oral and rectal candida colonisation is very common. In a previous study[10] we found that asymptomatic oral cavity or rectal candida colonisation is frequent among women with culture proven recurrent vulvovaginal candidiasis. However, a clinically evident oral thrush in these women was uncommon. In contrast, in the present study, 6 of 18 (33·3%) HIV-seropositive patients with symptomatic candidal vaginitis and oral specimens positive for candida had also clinically evident oral thrush. The persistent candida colonisation of the oral mucosa is considered a bad prognostic sign in HIV infected women.[2 6]

Non-albicans candida species are responsible for 10–15% of mycotic vulvovaginitis.[11 12] The prevalence of systemic infections caused by *Torulopsis glabrata* and other non-albicans species has increased during the last decade.[13 14] Accordingly, *T glabrata* is recognised as an increasing cause of recurrent vulvovaginal candidiasis.[11–14] It has been postulated that repeated antimycotic therapy, especially with imidazole derivatives, could have been responsible for the selection of non-albicans strains through a pressure selection mechanism.[14–16] Almost 60% of HIV-positive patients in this study reported a history of candida vaginitis in the year before the study and 30% of them had had recurrent infection treated mostly with imidazole derivatives. This may account for the high prevalence of non-albicans strains in the HIV-positive population. The high rate of *Torulopsis glabrata* or other non-albicans species as causal agents of recurrent vaginal infection in HIV-positive women should be taken into consideration from both the diagnostic and therapeutic viewpoint. In fact, the diagnosis of torulopsis vaginal infection on light microscopy may be difficult owing to the absence of hyphae and the characteristic spores. In addition, the infection is very difficult to eradicate since *Torulopsis glabrata* is usually less sensitive to the commonly used antimycotic agents, and resistance to the imidazole derivatives may also develop.[17] Boric acid used vaginally has also proven effective.[14]

In conclusion, this study has shown that in HIV-seropositive patients, vulvovaginal candidiasis is very common and is correlated with the immunological status of the host. These patients had higher frequencies of *Torulopsis glabrata* vaginal infection and are more prone to recurrence than HIV-negative controls.

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