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Single dose treatment of vaginal candidosis: Randomised comparison of amorolfine (50 mg and 100 mg) and clotrimazole (500 mg) in patients with vulvovaginal candidosis

Ein-Dosis-Behandlung der Vaginal-Candidose: Randomisierter Vergleich von Amorolfin (50 mg und 100 mg) und Clotrimazol (500 mg) bei Patientinnen mit Vaginal-Candidose

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Key words. Vaginal candidosis, amorolfine, clotrimazole, antimycotic chemotherapy, single dose treatment, topical treatment.

Schlüsselwörter. Vaginal-Candidose, Amorolfin, Clotrimazol, antimykotische Chemotherapie, Eindosis-Behandlung, topische Behandlung.

Summary. A double blind randomised comparative study of single dose treatment with amorolfine vaginal tablets (50 mg and 100 mg) and clotrimazole 500 mg monodose vaginal tablets (open labelled) was undertaken in patients with vaginal candidosis. Vaginitis was demonstrated by both a positive culture and positive findings on microscopic examination of a vaginal smear as well as by the presence of clinical symptoms. 118 patients seen over a 6 month period were randomly allocated to receive one 50 mg vaginal tablet of amorolfine (regimen A, 40 patients), a 100 mg vaginal tablet of amorolfine (regimen B, 38 patients) or a 500 mg tablet of clotrimazole (regimen C, 40 patients). At the assessment one week after the end of therapy the proportion of cured patients was 90% in group A, 94.7% in group B and 92.5% in group C. 4 patients (10%) in group A, 2 (5.2%) in group B and 3 (7.5%) in group C did not respond to the treatment. There was a significant association between *Candida glabrata* and treatment failure ($P < 0.001$) and *C. glabrata* and carrier state ($P < 0.01$). At the assessment 4 weeks after the end of therapy the proportion of cured patients was 80% in group A, 84.2% in group B and 67.5% in group C with a relapse rate of 10% (group A), 10.5% (group B) and 25% (group C). *C. glabrata* was significantly associated with non-effective overall treatment ($P< 0.05$). The relapse rate was significantly associated with positive culture results one week post therapy ($P< 0.05$). In terms of overall effectiveness, there were no significant differences between the three treatment groups.

Zusammenfassung. Es wurde eine randomisierte Doppelblind-Vergleichsstudie der Eindosisbehandlung mit Amorolfin-Vaginaltabletten (50 mg und 100 mg) und Clotrimazol-Einmalvaginaltabletten (500 mg) an Patientinnen mit Vaginalcandidose durchgeführt. Die Vaginitis war sowohl durch positive Kultur- und Mikroskopiebefunde an Vaginalabstrichen als auch durch die klinische Symptomatik gesichert. 118 Patientinnen wurden über eine Periode von 6 Monaten hinweg beobachtet und erhielten randomisiert entweder eine 50-mg-Amorolfin-Vaginaltablette (Regime A, 40 Patientinnen), eine 100-mg-Amorolfin-Vaginaltablette (Regime B, 38 Patientinnen), oder eine 500-mg-Clotrimazol-Tablette (Regime C, 40 Patientinnen). In der Kontrolluntersuchung eine Woche nach Therapie-Ende betrug der Anteil geheilter Patientinnen 90 % in

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Gruppe A, 94,7 % in Gruppe B und 92,5 % in Gruppe C. Vier Patientinnen (10 %) in Gruppe A, zwei (5,2 %) in Gruppe B und drei (7,5 %) in Gruppe C reagierten nicht auf die Behandlung. Eine signifikante Beziehung bestand zwischen *Candida glabrata*-Befall und Therapiemißerfolgen ($P < 0{,}001$) und *C. glabrata* und dem Keimträgerstatus ($P < 0{,}01$). In der Kontrolle vier Wochen nach Therapie-Ende betrug der Anteil der geheilten Patientinnen 80 % in Gruppe A, 84,2 % in Gruppe B und 67,5 % in Gruppe C mit einer Rückfallrate von 10 % bei Gruppe A, 10,5 % bei Gruppe B und 25 % bei Gruppe C. *C. glabrata* war signifikant korreliert mit dem Langzeit-Therapiemißerfolg ($P < 0{,}05$). Die Rückfallrate war signifikant korreliert mit positiven Kulturergebnissen bereits eine Woche nach Therapie ($P < 0{,}05$). Bezogen auf die Gesamtwirkung gab es keine signifikanten Unterschiede zwischen den drei Behandlungsgruppen.

Introduction

Vulvovaginal candidosis is one of the most common gynaecological complaints: its diagnosis is easy but its treatment can be a problem, and for the patients long therapeutic schedules can be cumbersome and often lead to non-compliance [1]. We report here the results of single dose treatment schedules comparing amorolfine and clotrimazole in 118 patients with vulvovaginal candidosis.

Amorolfine (RO-14-4767/002) is a new topical antifungal agent synthesised at F. Hoffmann-La Roche (Basle, Switzerland). This compound is a phenylpropylpiperidine (morpholine derivative), unrelated to the polyenes, imidazoles or allylamines, with high activity against yeasts and dermatophytes, observed both *in vitro* and *in vivo* [2, 3]. The compound has a fungistatic action based on the disturbance of ergosterol biosynthesis, inhibiting delta-14-reductase and delta-7-delta-8-isomerase, with final impairment of the cell membrane and subsequent cellular death [4]. Animal studies have shown amorolfine to be superior to imidazole derivatives and polyene antibiotics [2, 3]. In tolerability studies performed in healthy volunteers, no adverse events were observed following application of a 50 mg vaginal tablet of amorolfine for up to six consecutive days: negligible amounts of the compound were detected in plasma [5]. Three cream concentrations of amorolfine and the cream base, applied to intact and scarified skin and under occlusion were also well tolerated, and the compound was not detected in plasma, urine or faeces [5]. At present amorolfine is available in different galenic formulations, namely, vaginal tablets, vaginal ovules, alcoholic solutions, nail lacquer and cream containing different concentrations of the compound.

Clotrimazole (Bay b 5097, Canesten, Lotrimin, Mycosporin) was the earliest azole antifungal to be introduced for clinical use, and the pioneer drug used to demonstrate that short courses of high-dose azoles were effective in vaginal candidosis [6].

It has been shown that clotrimazole [6, 7] persists at inhibitory levels in vaginal secretions for up to 2–3 days after a single dose of 500 mg, and the trend now is towards shorter treatment courses with the aim of increasing patient compliance [8].

The aim of this dose finding study was to try to determine and compare the efficacy and tolerance of 50 mg and 100 mg vaginal tablets of amorolfine with 500 mg vaginal tablets of clotrimazole.

Material and methods

Medication

The trial was designed as a double blind randomised comparison of two doses of monodose amorolfine vaginal tablets (50 mg and 100 mg) with 500 mg single dose vaginal tablets of clotrimazole (open labelled). The vaginal tablets were inserted in the evening (in bed); no treatment was taken during menstruation. In the case of concomitant vulvitis or of balanitis of the partner, additional topical external treatment was permitted with cream. The cream was applied thinly on the affected area and rubbed in until complete healing was achieved (usually after 1 week). Amorolfine cream was applied once daily and clotrimazole cream twice daily.

Clinical assessment

One hundred and twenty non-pregnant women were included in the study, and all had symptoms such as vaginal burning, itching, reddening and leukorrhea, dysuria, dyspareunia, etc. At the initial visit patients were asked to grade their symptoms on a four-point scale (0 = not present; 1 = slight; 2 = moderate and 3 = severe). The physician's opinion of the intensity of leukorrhea, vaginitis and vulvitis was also noted. To evaluate tolerance adverse effects were recorded and rated according to severity as slight (1), moderate (2) and severe (3).

Microbiological assessment

Two specimens of vaginal discharge were taken with sterile swabs. One of the swabs was mixed with a drop of saline on a slide for direct wet-mount microscopy, and the other was plated immediately into (1) Columbia CNA agar (Difco, Detroit, Mich.) with 5% whole human blood; (2) Thayer-Martin selective medium (GC base, Difco, with 1% hemoglobin 5, Difco, and VCN inhibitor, (Oxoid, Bas-

ingstoke, Hampshire, UK)); (3) Sabouraud dextrose agar (Difco) with 0.5 mg of chloramphenicol/ml, and (4) trichomonas medium, No. 2 (Oxoid). All the plates were incubated at 37 °C in a CO_2 incubator.

The Minimum Inhibitory Concentration (MIC) values to amorolfine and clotrimazole were determined in Yeast Nitrogen Base liquid medium (Difco) to which glucose (5 g l^{-1}) was added using a macrodilution broth method (dilutions were two fold from < 0.05 to 50 µg ml^{-1}). A diluted inoculum (1:100) of a 24 hour culture in YNB broth was used.

Patients

All patients were clinically examined and had a detailed clinical history taken prior to treatment. Only patients with acute mycotic vaginitis (i.e. two episodes or more over the preceding year) confirmed by microscopy and culture were included in the study. Written consent was obtained from all patients. Pregnant women, women in whom pregnancy could not be excluded, patients with severe concomitant diseases or patients who had used topical or systemic antifungal treatment during the 4 weeks prior to the start of the trial were not included. Patients with *Trichomonas* infection, with *Neisseria gonorrhoeae* infection or with *Gardnerella vaginalis* vaginitis were also excluded from the study.

Evaluation

Patients were asked to return twice after treatment: one week after treatment (control 1) and one week after the end of menstruation following treatment (control 2), i.e. 4–5 weeks after the initial examination.

The clinical efficacy of the test drug was rated globally as: cured, improved, unchanged and deteriorated. The overall evaluation of the results was made at control 2 (4–5 weeks after the end of therapy) by correlating the clinical and mycological findings; the relapse rate was also documented.

'Cured' was defined by absence of clinical signs and symptoms, except for residual leukorrhoea, with negative mycological findings (microscopy and culture) or positive culture alone (carriers). All carriers had less than 9 yeast colonies per vaginal swab.

'Improved' was defined by improvement or amelioration of clinical symptoms with negative microscopy and culture.

'Treatment' failure was defined by improvement, stability or deterioration of clinical parameters with positive direct microscopy and/or positive culture. Patients with premature termination due to adverse effects were considered in this category for all follow-up visits or controls.

'Relapse' was defined as presence of clinical signs and symptoms of disease activity with positive microscopy and culture at control 2 in patients who were cured previously (at control 1).

Statistical analysis

Symptoms and clinical improvement were analysed using the Wilcoxon test to compare successive time points for each treatment group and using the Mann-Whitney U-test to compare results of treatment at controls 1 and 2. The χ^2-test was used to compare the mycological response (microscopy and culture) and tolerance (side effects).

Results

120 patients were included in the trial, randomised into one of the 3 parallel groups. Two patients in group B (see Table 1) dropped out of the study. Table 1 shows the demographic characteristics of patients, history and infecting organisms of the evaluated patients. Statistical examination of the patients randomised into one of the 3 groups showed the groups to be homogeneous at study entry.

Acceptability

All patients found the treatment easy, simple and cosmetically acceptable.

– Control 1 (one week post therapy):

The clinical and mycological response at the end of the treatment is recorded in Table 2. In group A (amorolfine 50 mg) there were 36 patients (90%) clinically cured, of whom 3 (7.5%) were carriers with negative microscopy and positive cultures of *C. glabrata*. There were 4 patients (10%) who did not respond to treatment. All had positive microscopy and *C. albicans* was grown from one and *C. glabrata* from three.

In group B (amorolfine 100 mg) there were 36 patients (94.7%) clinically cured of whom 4 (10.4%) were carriers with negative direct microscopy and positive cultures (*C. albicans* and *C. glabrata*, 1). There were 2 patients (5.2%) who did not respond to treatment. In both patients microscopy was positive and the cultures yielded *C. glabrata*.

In Group C (clotrimazole 500 mg) there were 37 patients (92.5%) clinically cured, of which 4 (10%) were carriers of *C. albicans*. Three patients (7.5%) were considered treatment failures, two with *C. glabrata* and one with *C. albicans*.

There was no significant difference in the clinical and mycological response when the three groups of patients were compared. The response to treatment according to the fungal species involved, showed in this short term assessment a significant association

Table 1. Demographic characteristics of patients and distribution of fungi isolated

Group	Amorolfine		Clotrimazole
Number of patients	A (50 mg) (*n* = 40)	B (100 mg) (*n* = 38)	C (500 mg) (*n* = 40)
Mean age (range) years	32.17 (21–52)	33.8 (19–54)	29 (24–46)
No. patients: previous fungal vaginitis	10	12	7
No. patients: first episode of fungal vaginitis	30	26	33
No. patients:			
No. predisposing factors, or underlying conditions	13	19	17
With concomitant diseases	2[a]	3[d]	1[g]
With predisposing factors	22[b]	14[e]	21[h]
With predisposing factors and underlying conditions	3[c]	2[f]	1[i]
C. albicans	34	33	36
C. glabrata	6	4	3
C. krusei	0	1	0
C. guilliermondii and *C. albicans*	0	0	1

(a) Diabetes (2). (b) I. U. D. (5); oral contraceptives (6); antibiotics (5); antibiotics and oral contraceptives (5); antibiotics and I. U. D. (1). (c) Hysterectomy and iron deficiency (1); hormonal treatment for sterility (1); mastopathy and oral contraceptives (1). (d) Toxid oil syndrome (1); diabetes (2). (e) Iron deficiency (1); I. U. D. (6); oral contraceptives (5); oral contraceptives and antibiotics (1); I. U. D. and antibiotics (1). (f) Hepatitis and antibiotics (1) I. U. D. and toxic oil syndrome (1). (g) Diabetes and rheumatoid arthritis (1). (h) Oral contraceptives (7); I. U. D. (8); Iron deficiency (1) Iron deficiency and oral contraceptives (1); Iron deficiency and I. U. D. (1); Oral contraceptives and antibiotics (3). (i) Toxic oil syndrome and I. U. D. (1).

Table 2. Results at control 1 (one week post therapy)

Group	Amorolfine		Clotrimazole
Number of patients	A (50 mg) *n* = 40 (%)	B (100 mg) *n* = 38 (%)	C (500 mg) *n* = 40 (%)
Clinical results			
Cure	36(90)	36(94.7)	37(92.5)
Improvement	0	0	0
Failure	4(10)	2(5.2)	3(7.5)
Mycological results			
Positive microscopy	4(10)	2(5.2)	3(7.5)
Positive culture	7(17.5)	6(15.7)	7(17.5)
Side effects	0	1(2.6)	1(2.5)

Table 3. Results at control 1 (one week post therapy)

	Failures (+ve culture)	Carriers*
C. albicans (*n* = 103)	2	6
C. glabrata (*n* = 13)	7	5
C. krusei (*n* = 1)	0	0
C. guilliermondii and *C. albicans* (*n* = 1)	0	0

* Carriers had negative microscopy and less than 9 yeast colonies per vaginal swab.

between *C. glabrata* and treatment failure (χ^2 test), $P < 0.001$) and also between *C. glabrata* and carrier state (χ^2 test, $P < 0.001$) [Table 3].

Sensitivity test. All the fungi isolated before treatment were sensitive to amorolfine and clotrimazole *in vitro* (Table 4). There was a difference, however, in the MIC values of *C. albicans* and *C. glabrata,* the former having lower values for both compounds than *C. glabrata.* For amorolfine 62% of strains of *C. albicans* were inhibited by less than 0.05 μg

Table 4. Fungistatic concentrations of clotrimazole and amorolfine

Yeast	MIC values (μg ml^{-1})	Number of isolates	
		Clotrimazole (*n* = 41 strains)	Amorolfine (*n* = 78 strains)
C. albicans	<0.05	23	42
	0.05	3	11
	0.1	6	5
	0.3	1	4
	0.4	2	2
	0.8	2	
	1.6		2
	3.2		1
C. glabrata	0.4		2
	0.8	3	7
	1.6		1
C. guilliermondii	0.05	1	
C. krusei	0.4		1

ml^{-1}, whereas none of the 10 isolates of *C. glabrata* were inhibited by less than 0.4 μg ml^{-1}, the majority (7) requiring 0.8 μg ml^{-1}. Similarly for clotrimazole 63% of strains of *C. albicans* were sensitive to less than 0.05 μg ml^{-1}, whilst the 3 *C. glabrata* strains required 0.8 μg ml^{-1} for inhibition. None of the strains isolated after treatment where shown to have developed resistance to either compound.

Side effects. One patient treated with amorolfine (group B, amorolfine 100 mg) had moderate irritation (burning and itching) that lasted half an hour when applying the amorolfine cream for the treatment of vulvitis; however the tolerance with the intravaginal medication was excellent.

In the clotrimazole treated group, one patient had moderate intravaginal irritation (itching for 12 hours following the application of the clotrimazole ovule); however, no side effects were noted when clotrimazole cream was used for the simultaneous treatment of her vulvitis (Table 2).

Patch tests were not performed because both patients refused consent.

– Control 2 (4 weeks post therapy):

The overall evaluation 4 weeks post therapy is recorded in Table 5. In group A (amorolfine 50 mg) there were 32 patients (80%) clinically cured; 26 patients had negative cultures (all had had *C. albicans* at baseline) and 6 patients (15%) were carriers with negative microscopy but positive cultures (*C. glabrata*, 3, and *C. albicans*, 3). There were 4 patients (10%) that relapsed: they all had positive microscopy and the infecting organism was *C. albicans*.

In group B (amorolfine 100 mg) there were 32 patients (84.2%) clinically cured of whom 25 had negative cultures (one had had *C. krusei* at baseline and 24 *C. albicans*). There were 7 asymptomatic carriers (18.4%) with negative microscopy from whom *C. albicans* was cultured. Four patients (10%) in this group relapsed (2 with *C. glabrata* and 2 with *C. albicans*).

In group C (clotrimazole 500 mg) there were 27 patients (67.5%) clinically cured, of whom 22 (55%) had negative cultures (one had had *C. glabrata* and 21 *C. albicans* at baseline). There were 5 asymptomatic carriers (12.5%) of *C. albicans*. Ten patients (25%) relapsed and in all the infecting organism was *C. albicans*.

There was no significant difference in the clinical and mycological response in the 3 groups of patients (Table 5). Table 6 shows the overall non-effective treatment for the 3 groups of patients, and includes the patients that did not respond to treatment at control 2; all of them required alternative antifungal treatment measures.

There was no significant difference in the overall non-effective treatment when the 3 groups were compared. In our population neither predisposing factors or underlying disease appeared to influence the results. Of 49 individuals without predisposing factors or underlying conditions, 12 patients did not respond to treatment or relapsed (3 in group A, 2 in group B and 7 in group C). Of 69 patients who hat predisposing factors or underlying conditions 15 did not respond to treatment or relapsed (5 in group A, 4 in group B and 6 patients in group C). With regard to the infecting species, *C. glabrata* was significantly associated with non-effective overall treatment (χ^2, $P < 0.05$). Of 103 cases infected with *C. albicans* there were only 18 where treatment was not effective compared to 9 of 13 with *C. glabrata*.

A separate analysis was made to see if the culture results (positive or negative) one week after the end of therapy had any influence on the outcome or relapse rate of vulvovaginal candidosis one month

Table 5. Results at control 2 (four week post therapy)

Group	Amorolfine		Clotrimazole
	A (50 mg) n= 40 (%)	B (100 mg) n = 38 (%)	C (500 mg) n = 40 (%)
Clinical results			
Cure	32(80)	32(84.2)	27(67.5)
Relapse	4(10)	4(10)	10(25)
Mycological results			
Positive microscopy	4(10)	4(10)	10(25)
Positive culture	10(25)	11(28.9)	15(37.5)

Table 6. Overall non-effective treatment (includes failures at control 1 and relapses at control 2)

Group	Amorolfine		Clotrimazole
	A (50 mg) n = 40 (%)	B (100 mg) n = 38 (%)	C (500 mg) n = 40 (%)
Control 1	4(10)	2(5.2)	3(7.5)
Control 2	4(10)	4(10.5)	10(25)
Total	8(20)	6(15.7)	13(32.5)

Table 7. Patients with positive cultures 4 weeks post therapy. Percentages are expressed in relation with the total number of patients (118) included at baseline

	Relapses (%)*	Carriers (%)**
C. albicans (n = 103 at baseline)	16(13.5)	15(12.7)
C. glabrata (n = 13 at baseline)	2(1.6)	3(2.6)

* All the patients had positive microscopy.
** All the patients had negative microscopy and less than 9 yeast colonies per vaginal swab in culture.

later. It was found that there was a significant association between positive culture results one week post treatment and the relapse rate one month later (χ^2, $P < 0.05$). Of 98 individuals with negative cultures at control 1, 13 relapsed (13.2%) as opposed to 11 patients with positive cultures (carriers) of whom 5 patients relapsed (45.4%).

Table 7 shows the fungal species distribution in the relapses and carriers 4 weeks after the end of the treatment.

Discussion

The results reported here suggest that single dose treatments of amorolfine (50 mg and 100 mg) are comparable in efficacy and tolerance to clotrimazole single dose (500 mg) for the management of acute vulvovaginal candidosis.

There are little published data available for amorolfine; most of the clinical studies being still in progress. A single dose treatment (50 mg amorolfine) vaginal tablets was shown to be more effective than 25 mg or 10 mg amorolfine tablets, achieving cure rates of 69.2%, 55.6% and 50% respectively, with good tolerances [5]. Another multicentre comparative study involving 259 patients and similar in design to our trial (single dose treatment comparing 50 and 100 mg vaginal tablets of clotrimazole) showed no significant difference between the three treatment groups regarding efficacy (cures were achieved in 74%, 77% and 71% respectively [5]. Our group of patients showed very similar responses, also with good tolerance. The morpholine antifungal derivatives offer the potential advantage of being an alternative choice of therapy for those patients known to have sensitivity to imidazole derivatives.

Vulvovaginal candidosis is very common in medical practice: it is estimated that approximately three quarters of all women suffer at least one attack of vaginal candidosis [9]. These patients require treatment, because spontaneous healing is uncommon, but it is estimated that only one third of the women use the medication as prescribed [10]. Therefore, short therapy is important for assuring patients compliance. In 1977, Odds [11] analysed statistically the data from large numbers of published trials of topical vaginal antifungals; the different antifungal formulations appeared not to influence mycological cure rates but a treatment duration of the order of 2 weeks was superior to shorter courses in terms of immediate mycological efficacy and prevention of recurrence. Gough [12], later analysing published reports, found that at least for miconazole, the total dose of antifungal used was as important as the duration of treatment in terms of mycological cures. In recent years, pharmaceutical companies have moved towards shorter treatment courses with high antifungal doses, leading to single-dose treatment alternatives.

It has been shown that topical imidazoles persist at inhibitory levels in the vaginal secretions for up to 2–3 days after a single dose. Clotrimazole [7, 13] isoconazole [14] and miconazole [15] thus persist as efficient levels locally, and are not systemically absorbed when applied topically in the vagina. Clotrimazole has an historical interest; it was the first topical imidazole found to be effective in a single high-dose for the treatment of vaginal candidosis [16]. In comparative studies it is, therefore, widely used as a control or reference drug.

In our patients the MIC values after therapy with either amorolfine or clotrimazole did not show any difference compared to pretreatment values, and therefore the development of secondary resistance to amorolfine after therapy does not seem to be a problem, although this should be confirmed with larger numbers of isolates. Despite the widespread use of imidazoles therapeutic failures due to the appearance of resistant organisms seems to be rare [17].

In the short term response there was a significant association between the causative organism being *C. glabrata* with failure to respond to treatment ($P< 0.001$) and also between *C. glabrata* and a persisting carrier state ($P < 0.01$). In our study the proportion of patients with overall non-effective treatment was also significantly higher in *C. glabrata* vulvovaginitis ($P< 0.05$). These strains also had higher MIC values than the other species tested *(C. albicans, C. krusei* and *C. guilliermondii)* both before and after treatment, suggesting an inherent or natural relative resistance and a correlation between clinical response and susceptibility *in vitro.*

From our data it would appear that *in vitro* tests could predict the clinical outcome, although it is well known that with imidazole and morpholine derivatives the antifungal MIC values vary depending on the test methods and experimental *in vitro* conditions [18]. Our results suggest that *C. glabrata* infections are more likely to respond unfavourably and this would correlate with *in vitro* natural resistance [18].

Other investigators have found higher MIC values for *C. glabrata* when compared to *C. albicans* [2], whilst others using different media and experimental conditions have not shown these differences [19].

It is well established that yeast carriage in the vagina is common in women with and without vaginal symptoms (18% and 11% respectively) [9]. Other surveys have reported a prevalence of 25% and 21%, respectively [20]. Our population showed a carrier state in 15% of the individuals (Table 7). Antifungal therapy of vulvovaginal candidosis

should achieve negative culture results since patients with positive cultures (even carriers with less than 9 yeast colonies per swab) seem to have a higher risk of relapsing ($P < 0.05$ in our patients with positive cultures one week after treatment).

More studies are needed with higher numbers of treated patients to define the role of amorolfine in the treatment of superficial fungal infections.

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References

1 Odds, F. C. (1988) *Candida and candidosis.* 2nd edn. London: Bailliere Tindall.
2 Polak, A. (1983) Antifungal activity *in vitro* of RO-14-4767/002, a phenylpropyl-morpholine. *Sabouraudia* **21,** 205–213.
3 Polak, A. & Dixon, D. M. (1987) Antifungal activity of amorolfine (RO-14-4767/002) *in vitro* and *in vivo.* In: *Fromtling, R. A. (ed.) Recent trends in the discovery, development and evaluation of antifungal agents,* pp. 555–573.
4 Müller, J., Polak, A. & Jaeger, R. (1987) The effects of the morpholine derivative amorolfine (RO-14-4767/002) on the ultrastructure of *Candida albicans. Mykosen* **30,** 528–540.
5 Rhode, E., Zang, M. & Hartmann, D. (1988) Preliminary clinical experience with RO-14-4767/002 (amorolfine) in superficial mycoses. In: Fromtling, R. A. (ed.) *Recent trends in the discovery, development and evaluation of antifungal agents,* pp. 575–582.
6 Lebherz, T., Guess, E. & Wolfson, N. (1985) Efficacy of single versus multiple – dose clotrimazole therapy in the management of vulvovaginal candidiasis. *Am. J. Obstet. Gynecol.* **152,** 965–968.
7 Mendling, W. & Plempel, M. (1982) Vaginal secretion levels after 6 days, 3 days and 1 day of treatment with 100, 200 and 500 mg vaginal tablets of clotrimazole and their therapeutic efficacy. *Chemotherapy* **28** (Suppl. 1), 43–47.
8 Masterton, G., Henderson, J. N., Napier, I. R. & Moffet, M. (1976) Vaginal candidosis. *Br. Med. J.* **I,** 712–713.
9 Berg, A. O., Heidrich, F. E., Hedayati, A. H. & Khami, M. A. (1984) Establishing the cause of genitourinary symptoms in women in a family practice. *JAMA* **251,** 620–624.
10 Robertson, W. H. (1985) The problem of patient compliance. *Am. J. Obstet. Gynecol.* **150,** 948–952.
11 Odds, F. C. (1977) Cure and relapse with antifungal therapy. *Proc. Roy. Soc. Med.* **70** (Suppl. 4), 24–28.
12 Gough, D. (1979) The influence of dosage and duration of administration of miconazole on the cure and relapse of candidal vaginitis. *Roy. Soc. Med. Int. Congr. Symp. Ser.* **7,** 15–20.
13 Lebherz, T., Guess, E. & Wolfson, N. (1985) Efficacy of single versus multiple – dose clotrimazole therapy in the management of vulvovaginal candidiasis. *Am. J. Obstet. Gynecol.* **152,** 965–968.
14 Tauber, U., Rach, P. & Lachnit, U. (1984) Persistence of isoconazole in vaginal secretion after single application. *Mykosen* **27,** 97–101.
15 Odds, F. C. & MacDonald, F. (1981) Persistence of miconazole in vaginal secretions after single applications of the antifungal: implications for the treatment of vaginal candidosis. *Br. J. Vener. Dis.* **57,** 400–401.
16 Milsom, I. & Forssman, L. (1982) Treatment of vaginal candidosis with a single 500 mg clotrimazole pessary. *Br. J. Vener. Dis.* **58,** 124–126.
17 Kobayashi, G. S. & Spitzer, E. D. (1989) Testing of organism for susceptibility to triazoles: is it justified? *Eur. J. Clin. Microbiol. Infect. Dis.* **8,** 387–389.
18 Odds, F. C. (1985) Laboratory test for the activity of imidazole and triazole antifungals agent *in vitro. Sem. Dermatol.* **4,** 260–270.
19 Shadomy, S., Espinel-Ingroff, A. & Kerkering, T. M. (1984) *In vitro* studies with four agents: BAY n 7133, Bifonazole (Bay h 4502), ICI 153066 and RO 14-4767/002. *J. Med. Vet. Mycol.* **22,** 7–15.
20 Bergman, J. J., Berg, A. O., Schneeweiss, R. & Heidrich, F. E. (1984) Clinical comparison of microscopic and culture technique in the diagnosis of candida vaginitis. *J. Pharm. Pract.* **18,** 549–552.