

Short-Term Topical Therapy of Experimental Tinea Pedis in Guinea Pigs with Lanconazole, a New Imidazole Antimycotic Agent

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Received 11 April 1995/Returned for modification 22 May 1995/Accepted 6 July 1995

The therapeutic efficacy of short-term treatment with a 1% cream of lanconazole, a new imidazole antimycotic agent, in comparison with that of a 1% cream of terbinafine was evaluated in the guinea pig model of tinea pedis. Each agent was topically applied once a day for 3 or 7 consecutive days, starting on day 10 postinfection, and a culture study was conducted on day 5 after the last treatment with each agent. The 1% cream of lanconazole was as highly effective as the 1% cream of terbinafine in terms of eradicating the fungi from the infected feet.

Lanconazole, (\pm)-(E)-[4-(2-chlorophenyl)-1,3-dithiolan-2-ylidene]-1-imidazolylacetonitrile, is a new imidazole antimycotic agent, and clinical trials with 1% cream and solution preparations completed in Japan demonstrated its therapeutic usefulness in the treatment of various dermatomycoses including tinea pedis, tinea corporis, and cutaneous candidiasis (9). Consistently, in preclinical studies the same preparations of lanconazole exhibited excellent therapeutic efficacy in guinea pig models of tinea pedis, tinea corporis, and cutaneous candidiasis (6-8). One percent lanconazole cream (Astat) is now commercially available in Japan.

Recently, as a major advance in antifungal chemotherapy, successful short-term treatment of dermatomycoses has been achieved by the use of topical terbinafine, an allylamine class of antifungal agent (2). In the present report, we address a comparative study on the short-term treatment of experimental tinea pedis with topical lanconazole and terbinafine.

Male Hartley guinea pigs (Japan SLC Inc., Shizuoka, Japan) weighing 500 to 700 g were used for the infection study. Animals were maintained in an air-conditioned room at $23 \pm 1^\circ\text{C}$ and were allowed access to feed and water ad libitum. The fungal strain used in the study was *Trichophyton mentagrophytes* TIMM 2789. The conidial suspension was prepared in sterile physiological saline containing 0.1% (wt/vol) Tween 80 from the fungus grown on Sabouraud's glucose agar slants for 2 weeks at 27°C . Following filtration through sterilized gauze to remove hyphal fragments and agar blocks, the final conidial suspension was adjusted to 10^8 conidia per ml for use as the inoculum.

Lanconazole was synthesized by Sumika Fine Chemical Co., Ltd. (Osaka, Japan), and its 1% cream was formulated by Tsumura & Co. (Tokyo, Japan) and is the same as the commercial product (Astat). The 1% cream of terbinafine (Lamisil; Sandoz Yakuhin Ltd., Tokyo, Japan) was used as a reference agent.

The procedures for infection were essentially identical to those described by Fujita and Matsuyama (4, 5). One side of a paper disk (1.5-mm thick by 8 mm in diameter; Advantec Toyo, Tokyo, Japan) was covered with a piece of aluminum foil, while

the other side was free to carry the inoculum suspension. The disk was immersed in 50 μl of the inoculum suspension and was then fixed on the planta pedis of animal feet with an adhesive elastic tape. The disk was removed on day 7 postinfection. To confirm the successful development of the infection, the skin of the infected sites was excised for microscopy on day 10 postinfection; this was followed by fixation, embedding in paraffin, and staining with hematoxylin and by the periodic acid-Schiff technique. Placebo or active formulations (0.1 g per locus) were topically applied to the whole soles of guinea pigs once a day for 3 or 7 consecutive days, starting on day 10 postinfection. Five days after the last treatment, the animals were sacrificed under ether anesthesia, and the skin of the infected sites was excised, washed quickly in 0.1% (wt/vol) benzalkonium chloride solution, and rinsed in physiological saline. The skin tissue from each planta pedis and the corresponding tarsus of all animals were cut into small blocks (about 2 by 2 mm). Ten skin blocks obtained from each part of the foot were implanted on a Sabouraud's glucose agar plate containing 20 IU of penicillin G per ml and 40 μg of streptomycin per ml and were cultured at 27°C for 14 days. The skin blocks yielding fungal growth were regarded as culture positive, and the foot (planta pedis plus tarsus) with more than one culture-positive skin block was considered fungus positive. In addition, the intensity of infection was given a score on the basis of the number of culture-positive skin blocks. Namely, a score of +10, +9, +8, +7, +6, +5, +4, +3, +2, +1, or 0 was given according to the corresponding number of culture-positive skin blocks of 10 skin blocks studied.

Statistical analyses for the rate of fungus-positive feet and for the average intensity of infection were assessed by Fisher's exact probability test and Mann-Whitney's U test, respectively. P values of <0.05 were considered significant.

Figure 1 shows photographs of skin sections of the infected site on day 10 postinfection. The fungal elements were frequently observed in the strata cornea of both the planta pedis and the tarsus, indicating that the infection was successfully established prior to treatment with the test agents. The results of the culture study are given in Table 1. In both the 7- and 3-day treatment studies, *T. mentagrophytes* was recovered from all feet of the untreated control groups of animals. In these animals, more than +9 and +7 of the average intensity of infection were obtained from the planta pedis and the tarsus,

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FIG. 1. Typical histological findings of skin tissues of the infected guinea pig foot. Sections were stained with hematoxylin and by the periodic acid-Schiff technique. Abundant hyphal elements invading into the stratum corneum were observed (arrows).

respectively. These results also indicate that the infection spread over the whole soles of the infected animals. In the vehicle-treated control group of animals, similar results were obtained. In the animals treated once daily with the 1% cream of lanconazole for 7 consecutive days, 7 of 10 infected feet became culture negative, and the rate of fungus-positive feet and the average intensity of infection were significantly lower than those for the untreated and placebo-treated control groups of animals. When the duration of treatment with the 1% cream of lanconazole was shortened to 3 consecutive days, the fungi were detected on 8 of 10 infected feet. The average intensity of infection in these animals, however, decreased significantly to +2.6 and +0.8 for the planta pedis and tarsus, respectively, compared with those for the untreated and vehicle-treated control groups of animals. Similarly, in the groups of animals treated with the 1% cream of terbinafine for 7 and 3 consecutive days, potent activity was also exhibited. There were no significant differences in the mycological pa-

rameters between the 1% cream of lanconazole-treated and the 1% cream of terbinafine-treated groups of animals.

Experimental tinea pedis in guinea pigs has been used for the evaluation of the activities of antifungal agents because the model is similar to naturally occurring tinea pedis in humans in terms of the histopathology, symptomatology, and chronicity and is more resistant to the antifungal therapies than the conventional tinea model produced on the backs of guinea pigs (1, 4, 5, 7, 8, 10). In this tinea pedis model, short-term treatment with the 1% cream of lanconazole exhibited almost the same potent activity as that with the 1% cream of terbinafine, and in particular, a 7-day treatment with each agent was long enough to obtain sufficient mycological cure. It has been reported that a 1-week course of treatment with the 1% cream of terbinafine is more effective than a 4-week course of treatment with the 1% cream of clotrimazole, an imidazole antifungal agent, in the treatment of naturally occurring tinea pedis in humans (3). Therefore, like terbinafine, lanconazole is expected to be highly effective in the short-term treatment of dermatomycoses in humans.

TABLE 1. Therapeutic efficacies of topically applied lanconazole and terbinafine in the guinea pig model of tinea pedis

Treatment	No. of culture-positive feet/ total no. of infected feet (%)	Average intensity of infection ^a	
		Planta pedis	Tarsus
3-day treatment			
Untreated control	10/10 (100)	+9.7	+7.8
Cream vehicle ^b	10/10 (100)	+8.8	+6.7
Lanconazole, 1% cream	8/10 (80)	+2.6 ^{c,d}	+0.8 ^{c,d}
Terbinafine, 1% cream	8/10 (80)	+2.6 ^{c,d}	+0.5 ^{c,d}
7-day treatment			
Untreated control	10/10 (100)	+9.9	+8.5
Cream vehicle ^b	10/10 (100)	+9.5	+8.5
Lanconazole, 1% cream	3/10 (30) ^{c,d}	+0.2 ^{c,d}	+0.1 ^{c,d}
Terbinafine, 1% cream	2/10 (20) ^{c,d}	+0.2 ^{c,d}	0 ^{c,d}

^a Significant differences from the comparable untreated control groups and the comparable cream vehicle groups are shown as $P < 0.01$ (c) and $P < 0.01$ (d) respectively. No significant differences were found in any of the parameters between the 1% lanconazole cream group and the 1% terbinafine cream group.

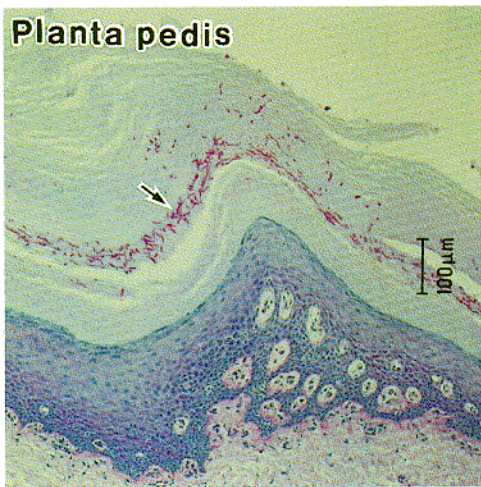
^b The cream vehicle of the 1% lanconazole cream.

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Planta pedis



Tarsus

