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Effect of AF-Anti Fungal Cream on Stratum Corneum and its Importance in Treating Cutaneous Mycoses

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Abstract

We have targeted the adhesion mechanism of fungal colonization to develop an effective siddha proprietary medicine-AF cream. The time tested siddha herbs like *Cassia alata* and *Azadirachta indica* were used to achieve total freedom from fungal infection. The findings show the uniqueness and the likely therapeutic value of AF cream in treating cutaneous fungal infection.

Keywords: Fungal infection; Adhesion; Stratum corneum cells; Tape stripping

Introduction

The incidence of cutaneous mycoses is growing in an alarming proportion all over the world [1,2]. The contribution of various lifestyle changes and associated disorders in the incidence and prevalence of cutaneous mycoses is quite significant. Diabetes mellitus, atopy, use, obsession and addiction towards wide spectrum of personal care and toiletry products do damage the cutaneous skin and thereby facilitate the colonization of various fungi.

The cutaneous mycoses is broadly classified under

Dermatophytoses (Ring worm)

Pityriasis

Candidiasis

The etiological agents that causes the above diseases belongs to diverse group of fungi viz., species under Trichophyton, Microsporum, Epidermatophyton, Malassezia, Candida [3,4].

Targeting the above species of fungi through a single antifungal agent is quite difficult. Abrogating the adhesion mechanism of fungi may offer a better solution to the above problem.

Way back in 1868, the British dermatologist Arthur Whitfield revolutionized the antifungal therapy by exploiting the concept of impairing the adhesion mechanism of fungi on cutaneous skin [5].

We in the present paper report the effect of AF cream, a proprietary siddha medicine (composed of the extracts of *Cassia alata* and *Azadirachta indica*) on stratum corneum and indicating its likely role in impairing the fungal adhesion [6,7].

Materials and Methods

Description of the product

AF cream is composed of

Cassia alata - 1%

Azadirachita indica - 1%

Cream base - QS

Preparation of cream suspension

For *in vitro* evaluation, 1% solution of AF cream and cream base (without herbal actives) were prepared in distilled water.

Evaluation of AF cream in removing stratum corneum

For this purpose we employed the modified method of Piérard, 1996 [8,9]. In brief high quality transparent cellophane tape was cut into 1 ½ inch length. The adhesive side of the tape was applied over the volar forearm region and gentle force was applied over the tape to gather all the loose stratum corneum cells. Six such tape stripping were taken from 10 volunteers. One tape stripping from each volunteer was used as untreated control. On the surface of second, third and fourth stripping of the all 10 volunteers, 1 mg/cm^2 AF cream was applied and incubated for 10, 20 and 30 minutes interval. The other two sets of tape strips were treated with AF cream base and water separately.

After incubation, the stripping were washed and stained using 0.1% methylene blue dye. The stained stripping were observed under microscope to evaluate the abundance and responsiveness of stratum corneum.

Various parameters such as extent of diffusion, extent of swell and extent of dissolution of stratum corneum were scored to ascertain the responsiveness of stratum corneum.

Corneosurfametry (CSM)

To score the effect of AF cream in removing stratum corneum in human volunteers as Minimal=+, Near complete=+++, Nil to negligible=–, we have performed CSM. In brief stratum corneum strippings (SCS) were prepared from the inner forearms of healthy volunteers. A solution of the test product or its neat formulation is sprayed over SCS placed in plastic tray. After a, given period of incubation at controlled temperature, the samples are thoroughly and gently rinsed in running tap water, air-dried, and stained for 3 min in a

toluidine blue-basic fuchsin solution. Thereafter, the samples are copiously rinsed with water and dried prior to perform colour quantification using reflectance colorimetry. If the test sample has the ability to remove the lipids, it would denaturate the corneocyte proteins, thus disclosing the chemical sites available for staining reactivity. A combined dotted and rimmed pattern is visible on corneocytes at the microscopic examination. Based on time of treatment SCS with the sample and the control the scoring was made and using this scale the *in vivo* findings were reconciled.

Results

Effect of AF cream in the removal of stratum corneum

The application of AF cream over 20 minutes has caused near complete removal of the loose stratum corneum cells. Whereas the AF cream base without CA+ complex (*Cassia alata* and *Azadirachta indica*) has affected the removal of stratum corneum only at minimal level (Table 1).

Treatment	Removal of stratum corneum versus Time in minutes		
	10	20	30
AF cream	+	+++	+++
AF cream Base	-	+	+
Water	-	-	+

Table 1: Effect of AF cream in the removal of Stratum Corneum. Minimal=+, Near complete=+++, Nil to negligible=-.

Responsiveness of stratum corneum to AF cream

Thirty minutes treatment of AF cream has caused high rate of dissolution of stratum corneum cells. Whereas AF cream base without CA+ actives did not affect the stratum corneum and so was water. When we used 1% NaOH solution, it had affected both partial diffusion and swelling of stratum corneum (Table 2).

Treatment	After 30 minutes		
	Extent of diffusion	Extent of swell	Extent of dissolution
AF cream	-	-	+++
AF cream Base	-	-	-
Water	-	+	-
NaOH (1% soln)	+++	+++	+

Table 2: Response of Stratum corneum to AF cream. Nil to no=-, Moderate=+, High=+++.

Discussion

The present study has clearly shown that AF cream is effective in removing all the senescent stratum corneum cells. This in turn would affect the availability of predominantly the damaged and desquamated stratum corneum cells for fungal colonization. The superficial cutaneous mycoses always limit their colonization to stratum corneum cells which are non-nucleated, dead, sclero protein deposition [10].

As early as in 1868, the British dermatologist Arthur Whitfield used the concept of impairing the adhesion of fungi over skin with the help of benzoic acid and salicylic acid and thus revolutionized the antifungal therapy [5].

We have used CA+ complex to challenge the fungal adhesion over the skin. Targeting the etiological cause although may serve better strategy in the treatment of cutaneous mycoses but when species of fungi belong to different genus are involved in the pathology, use of broad spectrum antifungal drugs are necessary. Most of the broad spectrum azole antifungal agents known to produce severe side effects and hence their use in superficial mycoses is quite limited. Only when the cutaneous mycoses become chronic and or extensive, the azole drugs are used [11].

Although a number of topical antifungal preparations with great specificity are available, in the era of emerging super bugs and drug resistant fungi, use of such antifungal drugs need special care and as far as possible, such preparations must be avoided.

The fungal adhesion inhibitors can offer great promise for treating cutaneous mycoses. Herbal actives can be a reliable candidate for the above purpose. In the present study we have used two medicinal herbs well documented in ancient siddha literature viz., *Cassia alata* and *Azadirachta indica* [6,7]. Ancient siddhars have identified several medicinal properties for the above plants including strong antifungal activity several thousand years ago. Our present study postulates that the above plants also may have strong adhesion block mechanism.

We have not compared the efficacy of AF cream with other antifungal preparation of polyene origin, allyl amine origin, triazole or imidazole origin. But still we assume in the light of above findings and siddha legacy to both the plants, AF cream will be quite effective.

References

1. Calado NB, Sousa FCD, Diniz MG, Fernandes ACS (2011) A 7-year survey of superficial and cutaneous mycoses in a public hospital in natal, northeast brazil. Brazilian Journal of Microbiology 42: 1296-1299.
2. Assaf RR, Weil ML (1996) The superficial mycoses. Dermatol Clin 14: 57-67.
3. Dekate S, Padhye S, Gautam A (2011) Identification and characterization of fungi causing superficial mycoses. Int J of Pharm and Life Sci 2: 782-786.
4. Thomas JW, Dennis MD (1996) Spectrum of Mycoses. Medical Microbiology. 4th edn. Chapter 75.
5. Whitfield A (1938) On the white reaction (white line) in dermatology. British Journal of Dermatology 50: 71-82.
6. Makinde AA, Igoli JO, Ama LTA, Shaibu SJ, Garba A (2007) Antimicrobial activity of Cassia alata. African Journal of Biotechnology 6: 1509-1510.
7. Natarajan V, Venugopal PV, Menon T (2003) Effect of azadirachta indica (neem) on the growth pattern of dermatophytes. Indian Journal of Medical Microbiology 21: 98-101.
8. Pierard GE, Goffi V, Pierard-Franchimont C (1994) Corneosurfametry: a predictive assessment of the interaction of personal care cleansing products with human stratum corneum. Dermatology 189: 152-156.
9. Pierard GE (1996) EEMCO guidance for the assessment of dry skin (xerosis) and ichthyosis: evaluation by stratum corneum strippings. Skin Res Technol 2: 3-11.
10. Kobayashi GS (1996) Disease of Mechanisms of Fungi. Medical Microbiology. 4th edn. Chapter 74.
11. Saag MS, Dismukes WE (1988) Azole antifungal agents: emphasis on new triazoles. Antimicrobial agents and chemotherapy 32: 1-8.