



STUDIES ON *IN VITRO* ANTIMYCOTIC POTENTIALS OF LOCAL AND INDUSTRIAL SOAPS ON VULVOVAGINAL *CANDIDA* SPECIES

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ABSTRACT: Soaps are known to possess antimicrobial properties, so it was hypothesised that soaps have *in vitro* antimycotic effects on *Candida* species associated with vulvo-vaginal candidiasis. A total of 129 strains of vulvo-vaginal and oral *Candida albicans* (43), *Candida glabrata* (52), *Candida pseudotropicalis* (19) and *Candida tropicalis* (15) obtained from endo-cervical / high vaginal swabs and oral rinses and swabs of non-diseased subjects of patients were bioassayed to determine their *in vitro* susceptibilities to 12 local, 18 industrial and 5 detergent soaps. None of the soaps had *in vitro* inhibitory activities on any of the *Candida* strains. In conclusion, commonly available local, industrial and detergent soaps assayed for in this study did not inhibit any of the test pathogenic *Candida* species *in vitro*, implicating that such soaps cannot be used in personal hygiene, disinfecting of contaminated undergarments nor for toilet purposes and therefore, cannot be of advantage as washing / germicidal agents or as topical cleansing agents in mycotic clinical conditions.

Keywords: antimycotic potentials, soaps, vulvovaginal *Candida*

INTRODUCTION

Vulvovaginal infection is the most common cause of gynaecological problems in sexually active women and few years ago it was considered as serious disease which may cause major health implications [1, 2]. Vulvovaginal candidiasis (VVC) or *Candida* vaginitis (vulvovaginal candidosis) is a common, worldwide significant mucosal infection, which accounts for 20-30% of gynaecological diseases observed in women, and affecting all strata of the society [3, 4]. Recurrent vulvovaginal candidiasis affects women worldwide [5], and the findings of [6] demonstrated that the cause of recurrent infections varies among individuals and ranges between strain maintenance, strain microevolution and strain replacement.

This cervical-vaginal infection, which currently implies life worsening, temporal indisposition, postoperative complications and even life threatening sepsis in patients hospitalised in Intensive Care Unit [2] is caused by opportunistic yeasts of the *Candida* genus. In spite of therapeutic advances in women's health, vulvovaginitis, VVC has been reported as one of the most frequent infections of the female genital tract with a high incidence, as well as increase in frequency. It causes extreme discomfort, such as vulvovaginal itching, severe burning, soreness, inflammation, whitish or whitish-gaudy, cottage cheese-like discharges, often with curd-like appearance. The patient also presents thick, fetid vaginal secretions with a granular appearance and an itchy, erythematous vulva; with the vagina becoming hyperaemic, while there may also be excoriation and dyspareunia [7]. Most *Candida* infections are treatable with antifungals [2] but sometimes, result in minimal complications and other feminine discomforts, which could be severe or fatal if left untreated in certain populations. Recurrent vulvo-vaginal candidiasis has also been attributed to re-infection due to contamination of underwears and other inner clothing of females [3, 8, 9]. This study therefore, investigates the *in vitro* inhibitory effects of commonly available soaps and detergents on *Candida* species, so as to serve as cleansing and washing agents of the vulvovaginal areas and underwear clothes, respectively, as a less conventional approach in the management of recurrent VVC.

MATERIALS AND METHODS

Collection of Clinical Specimens:

The vulvo-vaginal *Candida* strains used in this project were stock cultures obtained from Dr. Adenike Ogunshe's culture collections. The *Candida* strains were originally isolated from endocervical swabs (ECS) and high vaginal swabs (HVS) of patients presenting at the sexually transmissible infections (STI) clinic of the Department of Medical Microbiology & Parasitology, University College Hospital (UCH), Ibadan [4, 10].

Determination of anti-candidal activities of industrial and local soaps against *Candida* strains (modified agar well-diffusion method):

Eighteen industrial soaps, five detergents and 12 local soaps were bought from three major supermarkets and two herbal markets respectively within Ibadan metropolis. The names of the 18 industrial soaps were – Carat, Carex, Delta, Dettol, GIV, Halo antibacterial deodorant, London, Imperial Leather, Lux, Medisoft, Mekano, Meriko, Movate Savon, Premier antiseptic, Septol antiseptic & medicated, Tetmosol, Tura and Zee. The five detergent soaps were Omo multi active, Ariel, Sunlight, Bimbo and Klin.

The active ingredients of the industrial soaps were respectively listed as –GIV soap [*Cocos necitera* oil, *Elacsis guineensis* oil, sodium hydroxide, perfume, glycerin, rodo oil, titaniumdioxide, tetrasodium EDTA, water, CI 15880, CI 45100], Halo soap [sodium tallowate, sodium palm kernelate, water (Aqua), fragrance (Parfum), glycerin, sodium chloride, tetrasodium EDTA, tetrasodium etidronate, titanium dioxide CI 77891, menthol, trichlocarbon 1% w/w, CI 12490, limonene, butylphynyl, methylpropional, linalool, geraniol], Imperial Leather soap [soap base, water, glycerin, fragrance, stabiliser and colour], London soap [1.2% w/w mercuric iodide included as 3% potassium mercuric solution], Lux soap [sodium tallowate, sodium palmate, aqua, sodium palm kernelate, glycerin, paraffin, sodium sulphate, titanium dioxide, phosphoric acid, tetra sodium EDTA, etidronic acid, tocophenol acetate, disodium distyrylbiphenyl disulfonate, hexyl cinnamol, Geraniol, benzyl salicylate, butylphenyl, methylpropionol, coumarin, Limonene, CI 74160, Min TFM = 68%], Meriko soap [sodium tallowate, sodium cocoate, aqua, perfume, CI 74160, CI 12490, CI 77266], Tetmosol soap [5% monosulfiram B.P., sodium tallowate, *Citronella*], Tura soap [Triclosan, allantoin, vitamin E, sodium tallowate, sodium palm kernelate, aqua, perfume, CI 12940 (Pigment Red 5), CI 77266 (Carbon Black), CI 74160 (Pigment Blue 15)] and Zee soap [native black soap base, palm kernel oil, Shea butter (rich in natural vitamins), cocoa pod and palm bunch ash solution, camwood extract (*osun*), native honey, aloe vera, aqua, fragrance].

A modification of the Tagg *et al.* [11] method was used for the bioassay study to determine the *in vitro* inhibitory potentials of the various soaps. Sterile Mueller-Hinton agar was aseptically poured into sterile plates and allowed to set after which holes that were 6.0 mm in diameter were aseptically punched out of the agar plates. The agar surfaces were surface sterilised with Bunsen flame and allowed to cool before seeding the agar with the wound bacteria by streaking the entire surface of the Mueller-Hinton agar culture plates, after which the local and industrial soaps' solutions were separately dispensed into the holes and incubated at 35°C for 24-48 h. The modification was that the traditional and industrial soaps' solutions were incorporated into sterile semi-solid agar before dispensing into the agar wells, in order to avoid spreading of the solutions on the agar surface.

Demonstration of antagonism depends on the release into the assay media of diffusible inhibitory materials into the agar plates. Inhibitory zones surrounding the wound bacterial strains were noted and recorded in mm diameter, while zones that were less than 10.0 mm in diameter or absence of zones of inhibition were recorded as resistant or negative.



Plate 1: Local soaps used in the project study

RESULT

The *Candida glabrata* (52), *Candida albicans* (43), *Candida pseudotropicalis* (19) and *Candida tropicalis* (15) obtained from oral, endo-cervical and high vaginal swabs were not inhibited *in vitro* by any of the 12 local, 18 industrial (Carat, Carex, Delta, Dettol, GIV, Halo antibacterial deodorant, London, Imperial Leather, Lux, Medisoft, Meriko, Mekano, Movate Savon, Premier antiseptic, Septol antiseptic & medicated, Tetmosol, Tura and Zee) and 5 detergent (Omo multi active, Ariel, Sunlight, Bimbo and Klin) soaps (Tables 1-3).

Table 1: *In vitro* inhibitory potentials of industrial soaps on *Candida* species

| | DELTA | TETMOSOL | DETOL | SEPTOL | LUX | HALO | TURA | LONDON | IMPERIAL | ZEE | PREMIER | MEKIKO | MED SOFT | MOVATE | CAREX | GIV | MEKANO | CARAT |
|---------------------------------|-------|----------|-------|--------|-----|------|------|--------|----------|-----|---------|--------|----------|--------|-------|-----|--------|-------|
| <i>C. albicans</i> [43] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>Candida glabrata</i> [52] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. pseudotropicalis</i> [19] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. tropicalis</i> [15] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |

Table 2: *In vitro* inhibitory potentials of local soaps on *Candida* species

| | SP1 | SP2 | SP3 | SP4 | SP5 | SP6 | SP7 | SP8 | SP9 | SP10 | SP11 | SP12 |
|---------------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|------|------|------|
| <i>C. albicans</i> [43] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>Candida glabrata</i> [52] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. pseudotropicalis</i> [19] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. tropicalis</i> [15] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |

Table 3: *In vitro* inhibitory potentials of detergent soaps on *Candida* species

| | DTSP1 | DTSP2 | DTSP3 | DTSP4 | DTSP5 |
|---------------------------------|-------|-------|-------|-------|-------|
| <i>C. albicans</i> [43] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>Candida glabrata</i> [52] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. pseudotropicalis</i> [19] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| <i>C. tropicalis</i> [15] | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |

Keys: DTSP1 = Omo multi active; DTSP2 = Ariel; DTSP3 = Sunlight; DTSP4 = Bimbo; DTSP5 = Klin

DISCUSSION

No significant difference in the patterns of relatedness of the *Candida* strains was observed in this study, irrespective of which anatomical sites (oral cavity, endocervical and high vaginal regions) they were isolated from. Furthermore, it was also observed that a single host could be colonised with multiple strains of the same species at the same or different body sites, which indicates the dynamic processes of yeast colonisation in women [12]. Most of the *Candida* strains used in this study were however, from vulvo-vaginal swabs but still, the *Candida* species, *Candida albicans*, *Candida glabrata*, *Candida pseudotropicalis* and *Candida tropicalis* recovered from oral and vulvo-vaginal sources in this study were similar to those from same origin by previous studies.

The vaginal ecosystem harbours microbial flora that protects it from invading pathogens, especially those responsible for sexually transmitted diseases and urinary tract infections; however, a breach in the microbial flora can lead to vulvo-vaginal candidiasis. Recurrent vulvovaginal candidiasis (RVVC) affects women worldwide and approximately 75% of sexually active women suffer at least one episode of *Candida* vaginitis and up to 10% of them have recurrent episodes [5, 9, 13] associated with significant morbidity and may be notoriously difficult to manage [14]. Meanwhile, it has been reported that contamination and re-contamination of under wears may also be responsible for recurrence of vulvo-vaginal candidiasis in women. Therefore, reduction or total elimination of *Candida* contamination of such inner garments will ease the discomfort of candidiasis in women. Soaps play universal role in human hygiene; however, during the determination of *in vitro* inhibitory activities of 18 industrial soaps (Carat, Carex, Delta, Dettol, Giv, Halo, London, Imperial Leather, Lux, Med Soft, Meriko, Mekano, Movate, Premier, Septol, Tetmosol, Tura and Zee), 12 traditional (local/herbal) and six detergent soaps towards vulvo-vaginal *Candida* it was indicated that none of the industrial, local nor detergent soaps was inhibitory *in vitro*. Meanwhile, a number of other studies have reported the inhibitory potencies of various antimicrobial and non-antimicrobial soaps in clinical cases, especially as hand-washing and surgical agents [15, 16]. The study of Ogunshe *et al.* [10], evaluated the potentials of soaps, disinfectants and germicides as adjunct, topical cleansing agents in cases of candidiasis-associated vulvovaginal itching, and their findings indicated that out of a total of 39 medicated / toilet soaps, Crusader oil (100%), Meriko (95.0-100%), Tura (88.9%), Tetmosol (84.7%) and Aloe (68.4%) were the most-inhibitory (*in vitro*) soaps against *Candida* strains but the *Candida* strains were not susceptible to 33.3% of the soaps *in vitro*. However, apart from Meriko soap, the *in vitro* inhibitory activities of the remaining soaps varied within two-year interval, e.g., Meriko (100%: 95.0%), Tura (88.9% 8.2%) and Tetmosol (84.7% 26.7%).

A fact presented by the findings of this study can be that the batches of commonly obtainable soaps used in this study cannot be used for personal hygiene, like cleansing of human reproductive parts or washing of contaminated pants and other under wears, in cases of vulvo-vaginal candidiasis. It is very likely that the *Candida* strains were intrinsically resistant to the soaps; levels of chemical compositions (active ingredients) with inhibitory properties in the soaps were inappropriate or some of the soaps were adulterated. It must be strongly stressed that most of the soaps (just like other toilet / cosmetic products), and even especially clinical drugs in the country are either adulterated, fake or sub-standard.

This was rightly presented in the study of Ogunshe *et al.* [17], when it was reported that it is quite unfortunate that the menace of counterfeit and substandard drugs is being increasingly reported in developing countries like Nigeria. The importance of soap as an indispensable article in the household has not restrained the adulterators from making it a favourite object of their operations, and at the present day soap is only very rarely what it should be [18].

The study of Fuls *et al.* [16] indicated that non-antimicrobial soap was less active, so it can be deduced from the result findings of this study that genuine inclusion of certain well-known and very potent active ingredients like triclosan (a common ingredient in antibacterial soaps) in the Nigerian toilet and medicated soaps can increase the potentials of such soaps in hygiene and clinical cases, since it has been reported that antimicrobial soaps provide a greater microbial reduction than non-antimicrobial soaps. Triclosan is bacteriostatic at low concentrations but higher concentrations are bactericidal [19].

CONCLUSION AND RECOMMENDATION

The overall results of the present study indicated that the commonly available local, industrial and detergent soaps in the country did not inhibit any of the *Candida* species *in vitro*, which means that such soaps cannot be used in personal hygiene, disinfecting of contaminated undergarments nor for toilet purposes and therefore, cannot be of advantage as washing / germicidal agents or as topical cleansing agents in mycotic clinical conditions. Further studies to correlate the *in vitro* inhibitory activities reported in this study with same, similar or other soaps that are sold in other countries, as well as same soaps with different production batches are currently being carried out in our laboratories, in order to compare the *in vitro* inhibitory activities of soaps on *Candida* species associated with vulvo-vaginal candidiasis, as topical cleansing agents or washing agents in mycotic clinical conditions.

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Competing interests

The authors declare no competing interests

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