

Comparative Study of the Therapeutic Effect of Nigella Sativa Vaginal Suppository against Clotrimazole Vaginal Cream among Women with Candida Albicans Infection

Golsoum Sourazur^{1*}¹Department of Microbiology, Faculty of Microbiology, Islamic Azad University, Ahar Branch, Ahar, IranDOI: [10.36347/sajp.2022.v11i03.002](https://doi.org/10.36347/sajp.2022.v11i03.002)

| Received: 27.01.2022 | Accepted: 02.03.2022 | Published: 08.03.2022

*Corresponding author: Golsoum Sourazur

Department of Microbiology, Faculty of Microbiology, Islamic Azad University, Ahar Branch, Ahar, Iran

Abstract

Original Research Article

Objective and Background: Candida Albicans is responsible for 90% of vaginal fungal infections. This study aimed at comparison of the therapeutic effect of Nigella Sativa vaginal suppository against Clotrimazole vaginal cream among women with Candida Albicans vaginal infection. **Materials and Methods:** This interventional study is an analysis performed on 68 non-pregnant women suffering from Candida Albicans infection who referred to Alavi Hospital in Ardabil. They were divided into two equal groups of 34 people including Nigella Sativa (intervention group) and Clotrimazole vaginal cream (control group). Clinical and experimental symptoms and signs were observed and recorded by a questionnaire and form at the beginning of the study and after treatment. Treatment was performed in both groups for 7 nights. Data was analyzed by one-way ANOVA, t-test and McNemar Test. **Results:** The results of the study showed that the rate of improvement and treatment failure in the intervention and control groups was 86.30%-14.70% and 58.82%-41.18%, respectively. There was a significant difference between the two groups ($p < 0.002$). **Conclusion:** The present study showed that Nigella Sativa vaginal suppository provides a more effective therapeutic effect on Candida Albicans vaginal infection compared to Clotrimazole vaginal cream.

Keywords: Clotrimazole, Nigella Sativa, Candida Albicans infection.

Copyright © 2022 The Author(s): This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International License (CC BY-NC 4.0) which permits unrestricted use, distribution, and reproduction in any medium for non-commercial use provided the original author and source are credited.

INTRODUCTION

Candidiasis is the second most common vaginal infection and is usually caused by Candida Albicans. The infections caused by this microorganism occur in the lower genital tract and are found among 30-80% of women with asymptomatic or mild symptoms [1, 2]. In humans, the vagina is an organ that has a dynamic ecosystem containing some species of Lactobacillus. These microorganisms inhibit the growth of opportunistic pathogens by producing a series of inhibitors. Bacterial, trichomoniasis, and fungal infections commonly occur in women with severe Lactobacillus reduction. Topical application of drugs can cause proper growth of lactobacilli and thus reduce infection [3].

Candida Albicans is a dicotyledonous fungus in the form of blastospore that is in charge of transmitting and generating asymptomatic colonization. Also, Meselia (filamentous) as hyphae or pseudo-

hyphae that exacerbates the germination of blastospore produced by colonization and facilitates tissue invasion is necessary to attach to epithelial cells and has the highest degree of adhesion among the Candida Albicans species [2].

On the other hand, Candida Albicans is responsible for 85-95% of fungal infections of the vagina, and other types of Candida, such as Candida glabrata and Candida tropicalis, are responsible for the rest of the Candidal vulvovaginitis infections and tend towards medicinal resistance [4].

Common symptoms of Candidal vulvovaginitis include itching, burning of the urine (may occur when urine comes in contact with inflamed vestibule Epithelium). Cheese-like and dense scab-shaped, painful intercourse, abnormal vaginal discharge and erythema of the vulvar vaginal area [5] which can lead to physical and mental problems for the person

affected. The lesion may be eczematous, mild or severe, or accompanied by scabies and sores [6]. Some vulvar symptoms may occur, including edema, arrhythmias, and cracking.

In the classic case, the walls of the vagina are red and contain sticky, white and scan-shaped plaques [7].

Predisposing factors for Candidal vulvovaginitis infection include taking antibiotics, pregnancy, diabetes, estrogen therapy, immunosuppression, and obesity [3]. Vulvovaginitis is very common during the pregnancy and is more conventional in the last trimester of pregnancy due to a further decrease in vaginal pH. Due to high levels of estrogen and high concentrations of glycogen in vaginal discharges during pregnancy, the risk of Vulvovaginitis in these women is increased [7].

Various treatments have been proposed for the treatment of Candidal vulvovaginitis in the last two decades. The most common therapeutic agents are azoles, which are administered either topically in various concentrations and formulations or systemically. Clotrimazole can be pointed as one of the drugs used topically. Clotrimazole is a drug containing the azole group that is used to treat vaginal infections [8]. Besides, *Nigella Sativa* is a plant that in traditional medicine is called as an antimicrobial herbal medicine. Its scientific name is "*Nigella Sativa*" and its commercial name is "*Phytovagex*" belonging to the Ranunculaceae family. Its active ingredients include Carvacrol, Thymol, Bisimine, Anethole and Parasimine, which together with Thymoquinone cause antibacterial properties of *Nigella Sativa*, so that by changing the permeability of the cell membrane, it causes cell dysfunction and bacterial death [9, 10]. For example, Chaieb *et al.*, [11] report that the Thymoquinone available in *Nigella Sativa* have the power of inhibiting and preventing the formation of bacterial biofilms. On the other hand, *Nigella Sativa* increases the stimulation of the immune system, which has increased its effectiveness against various diseases [12]. This study aimed at comparison of the therapeutic effect of *Nigella Sativa* vaginal suppository against Clotrimazole vaginal cream among women with *Candida Allicans* vaginal infection.

METHODOLOGY

This work is an interventional-analytical study and was performed on 68 samples including non-pregnant women aged 15-45 years who referred to the gynecological clinic of Alavi Hospital in Ardabil for Candidal vaginitis (from February 2017 to February 2018).

Pregnant, breastfeeding and postmenopausal women, those having vaginal bleeding - sexual intercourse or vaginal douching in the last 24 hours,

those taking immunosuppressant drugs in the last 14 days, and women with a history of diabetes were excluded from the study. The rest of patients were examined for illness symptoms (e.g. itching, burning, etc.) and the patient's discharge was measured and recorded in terms of color, consistency, homogeneity and pH.

Then, after obtaining the patient's consent, sampling was performed using two sterile swabs. The first swab was sent to a microbiology laboratory for final diagnosis of *Candida Albicans* type in a tube containing normal saline for transfer to the Sabouraud Dextrose Agar (SDA) culture medium. The second swab was drawn on two slides. For the first slide, an expansion of vaginal secretions was prepared and by adding three drops of KOH, fish odor olfaction was examined; then, the samples were placed on the slide and the slide beneath the microscope was examined in terms of candida particularly in pseudo-hyphae form or germination yeast. The second slide was filled with 1-2 drops of normal saline solution under a microscope. In case of observation of key cells or flagellate parasites, they were excluded from the study due to the diagnosis of *Gardenerella* or *Trichomonas*.

Culture Method: First, the samples transferred to the laboratory were cultured for two days on SDA medium in a temperature of 25 degrees for separation of fungi. An amount of 0.5cc of human serum was mixed with some grown colony and then would be incubated in a temperature of 37 degrees for 3 hours. Next, the incubated samples were cultured in the Corn Meal Agar medium containing Tween 80 and then would be grown in a temperature of 25 degrees for 24 hours. The grown colony samples were observed under the microscope to examine presence of Chlamydo spores in Fungi. The *Candida Albicans* species were just included in the study.

In this study, the *Nigella Sativa* suppository was ordered by the drugstore to Isfahan Sabzdarou Pharmaceutical Company. Through the drugstore, 34 packages (each containing 7 pieces) were prepared from this company. Also, we obtained 50 grams of Clotrimazole vaginal cream made by Behvazan Iran through the drugstore.

The patients in the intervention group were provided with 7-pcs packages of *Nigella Sativa* suppository and each night, they were recommended to take 1 piece for 7 nights with the relevant applicator. Besides, the control group members were given Clotrimazole vaginal cream with the relevant applicator to be taken for 7 nights. All patients were provided with all the health points and they were asked to refer the clinic one week after completion of the treatment (*Candida Albicans* infection of all patients was confirmed by culture before treatment and culture was

once again undertaken after completion of the treatment) to evaluate the effect of drugs.

Data was analyzed using SPSS Statistical Software by one-way ANOVA descriptive test, and t-test and McNemar Test as the inferential tests.

RESULTS

In the present study (Figure 1) using one-way analysis, the highest incidence of *Candida Albicans* infection was in the age group of 21-29 years (with a mean age of 25.64) and the lowest percentage of infection was in the age group of 40-45 years (with an

average age 41 years), which were significant ($p < 0.005$) in terms of differences with other groups.

According to Table 1, the rate of improvement and failure of treatment in the intervention and control groups were 86.30%-14.70%, and 58.82%-41.18%, respectively. According to t-test, the difference was significant ($p < 0.002$).

According to Table 2 the intervention group was more sensitive and had less specificity compared to the control group on the clinical symptoms of *Candida Albicans*.

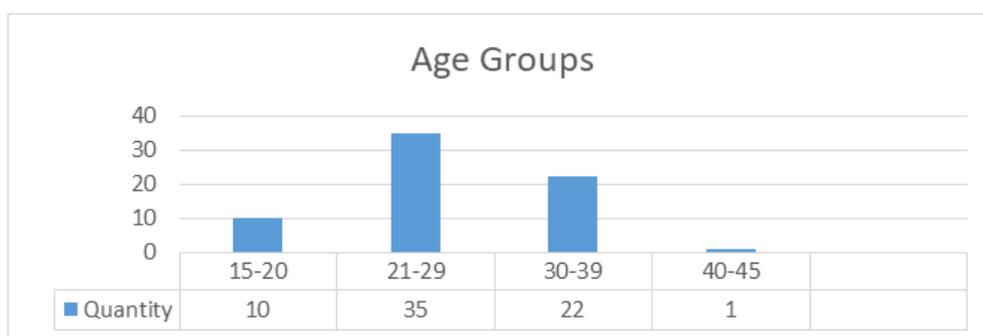


Figure 1: Distribution of frequency of patients with *Candida Albicans* separated by age groups (Alavi Hospital of Ardabil - from February 2017 to February 2018)

One-way analysis

Table 1: Comparison of the frequency distribution of Sabouraud Dextrose Agar (SDA) culture medium and wet smear before and after treatment in terms of Hyphae and Mycelium observation, between the control and intervention groups (Alavi Hospital of Ardabil - from February 2017 to February 2018)

Variable groups	The study duration					
	Before treatment (Positive)			The week after treatment (Negative)		
Hyphae and Mycelium observation	Clotrimazole vaginal cream Qty (%)	Nigella Sativa vaginal suppository Qty (%)	Result	Clotrimazole vaginal cream Qty (%)	Nigella Sativa vaginal suppository Qty (%)	Result
SDA culture medium	34 (100%)	34 (100%)	$p > 0.005$	14 (41.17%)	5 (14.70%)	$P < 0.001$
Wet smear	29 (91.17%)	29 (85.29%)	$p > 0.005$	18 (47%)	7 (20.58%)	$P < 0.001$
General result	$p > 0.005$			$P < 0.002$		

* Chi-square test - T test

Table 2: Comparison of sensitivity and specificity of the clinical signs between the two groups (Alavi Hospital of Ardabil - from February 2017 to February 2018)

Variable groups	Nigella Sativa vaginal suppository Qty (%)		Clotrimazole vaginal cream Qty (%)	
	Secretions	52.6	41.4	42.3
Foul-smelling discharge	64.9	51.3	52.6	56.2
Genital itching	72.5	42.2	63.5	44.4
Stimulation and irritation of the vulva	47.5	64.6	42.9	72.8
Inflammation	62.3	41.7	51.9	63.6
pH below 4.5	51.1	62.2	35.8	70.2
Edema of the vulva	51.1	62.2	35.8	70.2
Redness of the vulva	42.3	64.7	38.8	74.1

DISCUSSION AND CONCLUSION

The rate of improvement and failure of treatment in the intervention and control groups were 86.30%-14.70% and 58.82%-41.18%, respectively. There was a significant difference between the two groups ($p < 0.002$). In the study of Sovizi [13] entitled "comparison of the effect of Nigella Sativa suppository with Clotrimazole vaginal tablet in treatment of vaginal candidiasis", the success of treatment was 62% in women treated with Clotrimazole vaginal suppositories and 84% in the group of Nigella Sativa vaginal suppositories, which was a significant difference. This finding is in line with our results, which may be due to the active ingredient of Nigella Sativa. It has antifungal properties with compounds such as Thymol and Carvacrol. Nigella Sativa stimulates the immune system, which has increased its effectiveness against various diseases. In the study of Delirejh *et al.*, [12], it is reported that Nigella Sativa enhances the ratio of helper T cells to suppressor T cells and increases natural killer cells. In their study, increase in Phagocytosis index (Number of macrophages that have done Phagocytosis), and Phagocytosis speed were investigated. The number of phagocytosed particles was increased from 1-3 to 4-20 in each cell. The results of this study also showed that Nigella Sativa increases peripheral blood's lymphocytes and monocytes. However, the study of Sagafi *et al.*, [14] showed an improvement of 66% and 88% upon treatment by Clotrimazole vaginal cream and Nigella Sativa vaginal suppository, respectively, indicating an insignificant difference ($p = 0.013$). Their results were inconsistent with those of the present study in terms of significant differences. The reason for the difference between the two studies could be the research environment, duration of treatment and other variables.

In the present study, the intervention group was more sensitive and less specific than the control group on the clinical symptoms of Candida Albicans. In the literature review, Fard *et al.*, [15] showed a significant difference in improving genital itching, discharge, irritation and burning, and inflammation and redness of the vagina in the Nigella Sativa group compared to the placebo group ($P < 0.001$). Their results were approximately consistent with those of the present study, the level of awareness of individuals about abnormal genital signs and symptoms may be the reason for this alignment.

Nigella Sativa is a medicinal plant that is widely used in traditional medicine. On the other hand, according to other studies, this plant has antifungal and antibacterial effects, as well. Therefore, its use as a natural alternative treatment in infectious diseases is very important. According to the results of this study and the therapeutic effects of Nigella Sativa suppository in the treatment of Candida Albicans and its low side effects, its widespread use is recommended. The present study showed that Nigella Sativa vaginal suppository

has a more effective therapeutic effect on Candida Albicans vaginal infection compared to Clotrimazole vaginal cream.

ACKNOWLEDGEMENT

This research is a part of a master's thesis which has been approved under the number IR.AROMS.RFC.B95-96 in Ardabil University of Medical Sciences. We would like to thank all those who have contributed to this research.

Funding: No funding has been received for this project.

REFERENCES

1. Adalat Panahi, E., Rostampour, F., Poladi, G., & Rajajnejad, H. (2020). Evaluation of antifungal effects of Gyan Jashir (pangos fer ulace), and Barhang (PlantagoMigor L) against Candida albicans species resistant to Conazole flora in vitro. *Journal of Mashhad University of Medical Sciences*, 23(74), 44-52.
2. Shaer Mohammadi, M., & Nagizadeh, Z. (2016). *Comprehensive Diseases of Women's Diseases*. Second ed. Tehran: Andisheh Rafi Publications.
3. Asghari, B., & Jenabi Namin, M. (2017). *Henry Davidson Medical Microbiology*. McPherson RA, Pincus MR. 1st ed. Tehran: Artin Teb.
4. Mohammadi-Ghalehbin, B., Javanpour Heravi, H., Arzanlou, M., & Sarvi, M. (2017). Prevalence and Antibiotic Resistance Pattern of Candida spp; Isolated From Pregnant Women Referred to Health Centers in Ardabil, Iran. *Journal of Ardabil University of Medical Sciences*, 16(4), 409-421.
5. Esmaeilzadeh, S., Mahdavi, O. S., & Rahmani, Z. (2009). Frequency and etiology of vulvovaginal candidiasis in women referred to a gynecological center in Babol, Iran. *IJFS*, 3(2), 74-77.
6. Zaini, F., Mehbod, A. S. A., & Emami, M. (2009). *Comprehensive Medical Mycology*. 3rd ed. Tehran: Tehran University Press.
7. Valdan, M., Bouzari, B., Razzaghi, S., Jalilirad, A., & Nazem, S. (2008). *Danforth's Obstetrics and Gynecology*: Scott, J. R., Gibbs, R. S., Karlan, B., Haney, A., Beth, Y. 10th ed. Philadelphia: Wolters Kluwer.
8. Barousse, M. M., Van Der Pol, B. J., Fortenberry, D., Orr, D., & Fidel, P. L. (2004). Vaginal yeast colonisation, prevalence of vaginitis, and associated local immunity in adolescents. *Sexually transmitted infections*, 80(1), 48-53.
9. Bakathir, H. A., & Abbas, N. A. (2011). Detection of the antibacterial effect of nigella sativa ground seedswith water. *African Journal of Traditional, Complementary and Alternative Medicines*, 8(2), 159-164.
10. El-Dakhakhny, M., Madi, N. J., Lembert, N., & Ammon, H. P. T. (2002). Nigella sativa oil, nigellone and derived thymoquinone inhibit synthesis of 5-lipoxygenase products in

- polymorphonuclear leukocytes from rats. *Journal of ethnopharmacology*, 81(2), 161-164.
11. Chaieb, K., Kouidhi, B., Jrah, H., Mahdouani, K., & Bakhrouf, A. (2011). Antibacterial activity of Thymoquinone, an active principle of *Nigella sativa* and its potency to prevent bacterial biofilm formation. *BMC complementary and alternative medicine*, 11(1), 1-6.
 12. Delirejh, N., Morshedi, A., & Athari, S. S. (2010). Survey of the effect of powder *nigella sativa* (black seed) in increment of monocyte phagocytosis in quinea pig. *The Horizon of Medical Sciences*, 16(3), 55-63.
 13. Sovizi, B. (2007). Comparison of the effect of *Nigella Sativa* suppository with Clotrimazole vaginal tablet in treatment of vaginal candidiasis. Mashhad: Mashhad University of Medical Sciences.
 14. Sagafi, N., Rakhshandeh, H., & Sohadi, S. A. (2008). Comparative study of the therapeutic effect of cytochrome suppository suppository and vaginal suppository for the treatment of *Candida vaginitis*. Iranian Society of Physiology and Pharmacology, Mashhad University of Medical Sciences, 22, 18.
 15. Abbayan Fard F., Turk Zahrani, S., Akbarzadeh, A. R., & Mojib, F. (2012). Study of the Effect of Black Sea Capsules on the Treatment of Candidate Vaginitis in 2012. Tehran: Shahid Beheshti University of Medical Sciences.