

Studying the Airborne Fungi of some rooms in the internal sections of Mosul university campus and the Possibility of using Sage plants to control it

Maha Akram Al-Rejaboo*, Anfal Muayad Jalaluldeen

Department of Biology, University of Mosul, Mosel 41002, Iraq.

Correspondence: Maha Akram Al-Rejaboo. Department of Biology, University of Mosul, Mosel 41002, Iraq.

ABSTRACT

The purpose of this research was to determine the antifungal activity of sage plant (*Salvia officinalis* L.) alcoholic extract at varying concentrations (5, 10, 15 and 20 mg/ml) against a group of airborne fungus *in vitro* as a natural fungicide, which were isolated from indoor and chambers of Mosel University (*Alternaria*, *Acremonium*, *Aspergillus*, *Cladosporium*, *Drechslera*, *Fusarium*, *Nigrospora sphaerica*, *Penicillium*, *Rhizopus*, *Stemphylium*, *Tetereacoccosporium*, *Yeast*). Most of the fungi found in most rooms belonged to the genus, *Aspergillus*, *Penicillium*, *Cladosporium* and *Alternaria*. These fungus have been considered to be harmful fungi for humans, as they can cause allergies, dyspepsia, eye infections, skin injuries, and chronic bronchitis, etc. The minimum inhibitory concentrations of alcoholic extract against *Alternaria alternata*, *Aspergillus niger* and *penicillium* sp were 88, 76, 88% at the concentration of 20mg/ml compared with antifungal *Ketoconazol* which were 100%, 73% and 92% for the fungi *Alternaria alternata*, *Aspergillus niger* and *penicillium* sp; respectively at the concentration of 1.5 mg/ml. Based on the current results, alcoholic extract of *Salvia officinalis* exhibited good antifungal activity, and can potentially be a natural replacement of synthetic fungicides to control a number of significant fungal diseases.

Keywords: Antifungal activity, alcoholic extracts, Sage plant

Introduction

Indoor environments have essential roles in human health. The health hazards faced by humans from contaminated indoor environments comprise infections, allergies and toxicity. Shifts in modern lifestyles have also led to people spending less time outdoors, and more time in closed, air tight, energy efficient, environments, most of the times, indoor air pollution is caused

by hazardous biological agents and non-biological agents. In this regard, fungi are omnipresent, and widely distributed, posing the greatest danger to human health indoors [1].

Worldwide, the current trend is for many people to spend a major part of their lives indoors, either at home or at work [2, 3]. In such closed environments, unsatisfactory maintenance, inferior building design or occupant activities and behaviors frequently directly or indirectly create a condition known as "Sick Building Syndrome" (SBS), where occupants suffer adverse health effects that seem to be related to the long hours they spend within a building [4]. The complaints could be confined to just a particular room or be spread to an entire building, and relied only comes on leaving the premises [5]. Typical signs of SBS include pressure on the head, headaches and throbbing, and feelings of weariness.

In the past few years, much attention has been focused on plant extracts or essential oils, because of the natural compounds they contain. The biological activity exhibited by these compounds

Access this article online

Website: www.japer.in

E-ISSN: 2249-3379

How to cite this article: Maha Akram Al-Rejaboo, Anfal Muayad Jalaluldeen. Studying the Airborne Fungi of some rooms in the internal sections of Mosul University campus and the Possibility of using Sage Plants to Control it. *J Adv Pharm Edu Res* 2019;9(3):17-22.
Source of Support: Nil, Conflict of Interest: None declared.

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-Non Commercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

has enhanced the economic importance of these extracts and essential oils, leading to their increasing use in natural therapies and alternative medicine. Knowledge of these attempts to establish the effectiveness of these biologically active extracts and essential oils, may present an alternative to conventional pesticides. There are 236 genus and 7,133 species of plants presently recognized as belonging to the Lamiaceae family, which has an extensive geographic distribution globally [6]. The *Salvia* L. genus (Lamiaceae), which is known in Turkey as sage, is a significantly promising medicinal plant that is widely cultivated around the world. Due to their rich content of essential oils, *Salvia* species are aromatic plants that are commonly used not only as spices and traditional remedies, but also in the cosmetics industry and perfume. *Salvia* has been commonly consumed among the population as a bloating remedy, cold remedy, diuretic, stomach relaxant, and tonic drink [7, 8].

More than 95 *Salvia* species in the nation have been reported in recent research [9]. Plants in the Family of Lamiaceae and, to a lower degree, in the genus *Salvia*, include flavonoids, phenolics and vital oils and quinonoids [10]. They also contain flavonoids [11-13]. Oils and extracts of these crops have elevated biological activity thanks to their content of terpenoids and phenolic compounds. Studies in the literature reported that the *Salvia* species have had antibacterial properties [14], antifeedant [15], antioxidant [16], cytotoxic [17], antiviral [18], antifungal [19, 20], and antimicrobial effects [21, 22]. The antimicrobial impacts of *S. cryptantha*, an endemic species, have also been studied. *Salvia officinalis* L. is a perennial and perennial subshrub, made up of grayish, woody and blue to purplish flowers, with a wise, sage garden or wise frequency. It comes from the Mediterranean region, but is presently grown in numerous nations around the globe in many nations globally [20]. The aerial components are grown and only dried to achieve unstable oil. The unstable petroleum content can differ from 0.2% to 0.9% of the raw material from 0.5% to 2.65% for dry matter depending on the way it is sown and the environment. The seed should start at the peak of the volatile oil content before the flowering stage [7]. The composition and antifungal activity of *Salvia* was explored in this research. The alcoholic officinal is obtained from flowers and leaves in the inner departments of Mosul University against air fungus species. In this study, this activity was compared with ketoconazole antifungal agents in broth micro dilution assays.

Materials and Methods:

The samples were gathered from air of bed rooms and kitchens from different residential complexes of the university of Mosul, such as: Al-Zahraa complex with its first and second parts, Al-Qadisseyah campus with its third and sixteenth parts by taking material Petri dishes on local potato dextrose agar (PDA) and add antibiotic streptomycin, open it for half an hour, and the petri dishes were incubated at 28 °C for seven days with continuous supervision.

The fungi were diagnosed formally according to the fungi colony color, stature, form and danger, and microscopic diagnosis was used, the results were registered based on the classification keys [23-27].

The following dispersive locates were used for diagnosis: 25 percent Glycerol Nitrate Agar (G 25 percent N) Czapek yeast mineral extract agar (CYA), Malt extract agar (MEA), which were incubated on three temperatures: 5 °C, 25 °C, 37 °C for seven days, the results were registered, and they were compared with the previous classification keys for diagnosis.

Preparing the Alcoholic Extract for the Sage Plant:

The alcoholic extract was prepared by soak powder of dry leaves for the sage plant in ethanol with concentration 99%, after mowing it in Homogenizer set, then saved in closed dispenser in 4 °C, 8 °C for three days. The extract was transpiring by many layers of gauze, then submitted to the operation of centrifuge 2000 round/ minutes for losing the plant rests, the extracts were dried, then a certain weight was taken from it and melted in (Dimethyl sulphoxide) (DMSO), so there was a standard dissoluble regarding its concentration estimated by mg/ml, and purified by using micro filter paper (pore size 0.25 mm), then the concentrations (5, 10, 15, 20) mg/ml were prepared from the extract with locale (PDA) by using the alleviating equation ($N1V1 = N2V2$) for getting the minimum inhibition concentration, as well as the comparison processing that material Petri dish on the planting locale (PDA) without and addition, the tested the fungi isolations sensitivity toward this extract by plant discs with diameter of 6 mm from fungi colony in seven days, and then three repetitions were used for each concentration, and the dishes were incubated on 2 ± 28 °C for seven days, then the results were registered [28, 29].

Results and Discussion:

Random samples were collected from the air at Al Zahraa and Al Qadisyyah campus, which is located inside Mosel University, located on the campus where many of the fungi were found on the Al Zahraa chambers where air was isolated as shown in Table (1) and Table (2).

Table 1: Studies on airborne fungi from the different aria inside Al Zahraa campus.

Location	Predominant Fungi
Section (1) Room (4) First floor	<i>Tetracosporium paxianum</i> , <i>Aspergillus niger</i> , <i>Cladosporium</i> sp., <i>Penicillium</i> sp., <i>Penicillium spinulosum</i> <i>Cladosporium varibile</i>
Section (1) Room (14) First floor	<i>Penicillium spinulosum</i> , <i>Tetracosporium paxianum</i> <i>Aspergillus niger</i> , <i>Cladosporium cucumerinum</i> , <i>Alternaria sonchi</i> , <i>Alternaria alternata</i>
Section (1) Room (23) Second floor	<i>Tetracosporium paxianum</i> , <i>Alternaria alternata</i> <i>Aspergillus niger</i> .

Section (1) Room (33) Third floor	<i>Alternaria sonchi</i> , <i>Cladosporium oxysporum</i> <i>Penicillium spinulosum</i>
Section (1) Room (31) Third floor	<i>Tetracoccosporium</i> , <i>Alternaria</i> , <i>Penicillium sp.</i> <i>Aspergillus niger</i> , Yeast.
Section (1) Room (4) First floor	<i>Tetracoccosporium paxianum</i> , <i>Aspergillus niger</i> , <i>Cladosporium sp.</i> , <i>Penicillium sp.</i> , <i>Penicillium spinulosum</i> , <i>Cladosporium varibile</i>

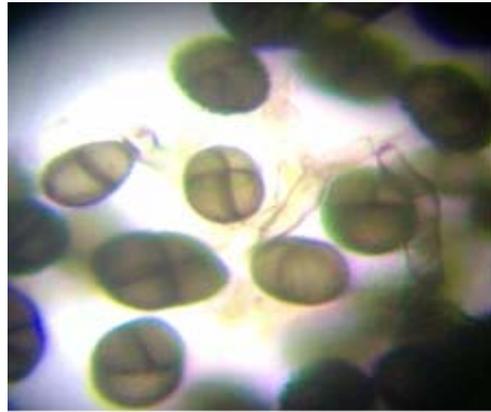


Table 2: Studies on airborne fungi from the different aria inside Al Qadisiyah campus

Location	Predominant Fungi
Section (3) Room (B) Apartment (9)	<i>Penicillium sp.</i> , <i>Cladosporium</i> <i>Nigrospora sphaerica</i> .
Section (16) Room Kitchen Apartment (8)	<i>Alternaria sonchi</i> Yeast
Section (16) Room (B) Apartment (8)	<i>Alternaria sonchi</i> , <i>Alternaria dianthi</i>
Section (16) Room Kitchen Apartment (9)	<i>Penicillium sp.</i> , <i>Cladosporium sp.</i> <i>Alternaria sp.</i> , <i>Cladosporium spongiosum</i> .Yeast
Section (16) Room (D) Apartment (7)	<i>Fusarium sp.</i> , <i>Drechslera biseptata</i> <i>Alternaria alternata</i>
Section (16) Room (C) Apartment (7)	<i>Penicillium sp.</i> , <i>Alternaria sonchi</i> , <i>Alternaria raphani</i> <i>Cladosporium oxysporum</i> , <i>Cladosporium varibile</i> .
Section (16) Room (A) Apartment (9)	<i>Drechslera cochliobolus</i> , <i>victoriae</i> , <i>Alternaria dianthi</i> <i>Cladosporium cucumerinum</i> , <i>Alternaria snochi</i> , <i>Aspergillus niger</i> , <i>Cladosporium herbarum</i> , <i>Penicillium commune</i> , <i>Penicillium spinulosum</i> . <i>Penicillium restrictum</i>
Section (16) Room (C) Apartment (9)	<i>Alternaria dianthicola</i> , <i>Alternaria raphani</i> , <i>Acremonium blochii</i> , <i>Stemphylium pleospora</i> . Yeast.
Section (16) Room (B) Apartment (9)	<i>Rhizopus stolouifer</i> , <i>Drechslera hawaiiensis</i> . <i>Penicillium variabile</i> .
Section (3) Room (D) Apartment (3)	<i>Alternaria alternata</i> , <i>Stemphylium vesicarium</i> <i>Mycelia sterilia</i>





Figure 1: Petri dish contaminated with fungi isolated from (Al Zahraa and Al Qadisiyah campus).

It has been shown through the results that isolated fungi cause disease for humans, for example *Alternaria alternata* fungus causes skin lesions and inflammation of the internal eye after eye surgery; also, the genus *Alternaria dianthicola* causes injuries under the skin, as for fungus *Cladosporium herbarum*, it has been recorded that this type might cause the occurrence of inflammation of keratinized energy, and *Penicillium spinulosum* fungus it caused neuromuscular inflammation with bronchitis, and the fungus *Aspergillus niger* caused external ear injuries [30, 31].

It was also noted that *Drechslera hawaiiensis* fungus causes sinusitis as well as pulmonary and other diseases, skin dermatitis, internal and technical eye inflammation.

Rhizopus stolonifer might cause a satisfactory condition in the eye, brain and nose. This indicated that the presence of fungi in the air of the rooms, especially those that are inhabited for long periods, such as the bedrooms and the kitchen might cause diseases in the population, such as allergies and skin infections.

Table 3 shows the effect of the sage plant (alcoholic extract) against the fungi found in the internal chambers, and also the effect of the antifungal *Ketoconazol* on the rate of fungal colonies (cm.).

Table 3: The effect of the sage plant alcoholic extract against the fungi found the antifungal Ketoconazol on the rate of fungal colonies (mg/ml.)

Fungus	Concentration of sage plant (cm.)				Concentration of antifungal Ketoconazole (cm.)				Control
	5	10	15	20	0.5	1	1.5	2	
<i>Alternaria alternata</i>	2	1.5	1.1	0.8	0	0	0	0	7
<i>Aspergillus niger</i>	4	3	2.5	1.8	5.3	3	2.1	0	7.8
<i>Penicillium sp.</i>	2	1.5	1	0.8	0.8	0.8	0.5	0	7

In table (3), the salad plant was efficient against fungus and inherited a concentration similar to that of certain other antifungal agents from the development of all bacteria tested dose-dependently. It was shown that the growth of fungi when the sage plant alcoholic extract was used compared with

antifungal Ketoconazole that the growth of the fungal *Alternaria alternata* in concentrations of 5,10,15 and 20 mg/ml was about 2,1.5,1.1 and 0.8 cm; respectively. While the fungus *Aspergillus niger* and *Penicillium sp.* had a growth rate of 4,3,2.5,1.8cm; respectively on concentrations of 5,10,15 and 20 mg/ml. From these results, it was found that the increase of the extract percentage led to the decrease of the growth rate of fungi, and this result agreed with [32], that the extract of *Salvia officinalis* showed a good antifungal activity, and could serve as a natural alternative to synthetic fungicides for the control of some important fungal diseases. Table (4) shows that the percentage of the effect of the sage plant extract and the antifungal ketoconazole on fungus colonies, the fungus *Alternaria alternata* was inhibited by 50% at the concentration of 5 mg/ml while it was 78%, 84% and 88% at concentrations of 10%, 15 and 20 mg/ml; respectively.

And, the antifungal Ketoconazole completely inhibited the fungus at all concentrations in combination with the control, this was agreed with [33]. Every extract had a powerful antifungal impact on the fungal culture; also, [34] showed important fungal antifungal inhibition in the range of 5-10 mg / ml of Bioassays with *Salvia external lachnocalyx*.

Table 4: The percentage inhibition of the sage plant alcoholic extract, and the antifungal ketoconazole on fungus colonies.

Fungus	Percentage inhibition								Control %
	inhibition of sage plant extract				of Ketoconazole%				
	5	10	15	20	0.5	1	1.5	2	
<i>Penicillium sp.</i>	50	78	85	88	88	88	92	100	0
<i>Alternaria alternata</i>	50	78	84	88	100	100	100	100	0
<i>Aspergillus niger</i>	48	61	67	76	32	61	73	100	0

The fungus *Aspergillus niger* was inhibited by 48% at the concentration of 5 mg/ml, and about 61%, 67% and 76% for the concentrations of 10, 15 and 20 mg/ml, while the antifungal Ketoconazole inhibited the fungi by 32% at 0.5% concentration, and 61% and 73% at concentration 1 and 1.5 mg/ml; respectively, while the concentration of 2 mg/ml completely reduced the growth of fungus compared to the control sample. For the fungus *penicillium sp.*, the extract reduced the growth about 50% at the concentration of 5 mg/ml; and 78% , 85% and 88% at the concentrations of 10,15 20 mg/ml; respectively, while the effect of the fungal antagonist ketoconazole was 88% at the concentration of 0.5 ,1 mg/ml, and 92% at 1.5 mg/ml concentration, while completely inhibited the fungal at the concentration of 2 mg/ml compared with the control sample, from this result, it was noted that there was an effect of sage plant on these fungi and this was consistent with [35] and [36] that sage plant had a clear inhibitory effect on fungi.

It was also noted that the sage plant can be used to sterilize the room air by bulling the sage leave or used as sprays used for

human compared with the side effect of using antagonistic fungi [37].

Conclusions:

The findings favored the antifungal activity of the *Salvia officinalis* alcoholic extract. More investigation of the antifungal components of the salvia of alcoholic extract, and the purification of these products would help to develop fresh natural antifungal medicines for resistant strains of the fungi that could be supplemented by accessible synthetic agents such as antifungal agents (*Alternaria alternata*, *Aspergillus niger* and *penicillium sp.*).

References

1. A.A. Haleem Khan, S. Mohan Karuppaiyl (2012) Fungal Pollution of Indoor Environments and its management; Saudi Journal of Biological Sciences doi: [http:// dx.doi.org /10.1016/j. sjbs. 2012.06.002](http://dx.doi.org/10.1016/j.sjbs.2012.06.002)
2. Chao, H. J., Schwartz, J., Milton, D. K., Burge, H. A., (2003). The work environment and workers health in four large office buildings. Environmental Health Perspectives, 111 (9), 1242-1248.
3. Molhave, L., (2011). Sick building syndrome. Encyclopedia of Environmental Health, 61-67.
4. Ebbehøj, N. E., Hansen, M. O., Sigsgaard, T., Larsen, L., (2002). Building-related symptoms and molds: a two-step intervention study. Indoor Air, 12, 273-277.
5. Bakke, J. V., Norbäck, D., Wieslander, G., Hollund, B. E., Florvaag, E., Haugen, E. N., Moen, B. E., 2008. Symptoms, complaints, ocular and nasal physiological signs in university staff in relation to indoor environment - temperature and gender interactions. Indoor Air, 18 (2), 131-143.
6. Harley, R.M., Atkins, S., Budantsev, A., Cantino, P.D., Conn, B.J., Grayer, R., Harley, M.M., De Kok, R., Krestovskaja, T., Morales, R., Paton, A.J., Ryding, O., Upson, T. (2004) Labiatae. In: Kubitzki, K. (ed.) The families and genera of vascular plants. vol. 7, Springer- Verlag, Berlin, 167-275.
7. Bayram, E. (2001) A Study on selecting suitable types of the Anatolia sage (*Salvia fruticosa* Mill.) in the Flora of Western Anatolia. Turk. J. Agric. For. 25 (6), pp: 351-357.
8. Amiri, H. (2007) Quantative and qualative changes of essential oil of *Salvia bracteata* Bank et Sol. in different growth stages. DARU. 15 (2), pp:79- 82.
9. Celep, F., Dogan, M., A. (2009) A new record for the flora of Turkey: *Salvia viscose* Jacq. (Labiatae). Turk J Bot. 32, 57-60.
10. Ulubelen, A., Miski, M., Neuman, P. and Mabry, T.J. (1979) Flavonoids of *Salvia tomentosa* (Labiatae). Journal of Natural Products. 42(3), 261-263.
11. Durling, N.E., Catchpole, O.J., Grey, J.B., Webby, R.F., Mitchell, K.A., Foo, L.Y. and Perry, N.B. (2007)

- Extraction of phenolics and essential oil from dried sage (*Salvia officinalis*) using ethanol-water mixtures. *Food Chemistry*. 101 (4), pp: 1417-1424.
12. Bisio, A., Damonte, G., Fraternali, D., Giacomelli, E., Salis, A., Romussi, G., Cafaggi, S., Ricci, D. and Tommasi, N.D. (2011) Phytotoxic clerodane diterpenes from *Salvia miniata* Fernald (Lamiaceae). *Phytochemistry*. 72 (2-3), 265-275.
 13. Al-Qudah, M., Al-Jaber, H., Zarga, M.H.A. and Orabi, S.T.A. (2014) Flavonoid and phenolic compounds from *Salvia palaestina* L., growing wild in Jordan and their antioxidant activities. *Phytochemistry*. 99, 115-120.
 14. Kawahara, N., Tamura, T., Mayumi, I., Hosoe, T., Kawai, K., Sekita, S., Satake, M. and Goda, Y. (2004) Diterpenoid glucosides from *Salvia greggii*. *Phytochemistry*. 65 (18), 2577-2581.
 15. Fraga, B.M., Diaz, C.E., Guadano, A. and Gonzalez-Coloma, A. (2005) Diterpenes from *Salvia broussonetii* transformed roots and their insecticidal activity. *J. Agric. Food Chem*. 53 (13), 5200-5206.
 16. Lakkhal, H., Ghorab, H., Chibani, S., Kabouche, A., Semra, Z., Smati, F., Abuhamdah, S. and Kabouche, Z. (2013) Chemical composition and biological activities of the essential oil of *Salvia officinalis* from Batna (Algeria). *Der Pharmacia Lettre*. 5(3), 310-314.
 17. Lee, W.Y.W., Cheung, C.C.M., Liu, K.W.K., Fung, K.P., Wong, J., Lai, P.B.S. and Yeung, J.H.K. (2010) Cytotoxic effects of tanshinones from *Salvia miltiorrhiza* on doxorubicin-resistant human liver cancer cells. *J. Nat. Prod*. 73 (5), 854-859.
 18. Tada, M., Okuno, K., Chiba, K., Ohnishi, E. and Yoshii, T. (1994) Antiviral diterpenes from *Salvia officinalis*. *Phytochemistry*. 35 (2), pp: 539-541.
 19. Jassbi, A.R., Mehrdad, M., Eghtesadi, F., Ebrahimi, S.N. and Baldwin, I.T. (2006) Novel rearranged abietane diterpenoids from the roots of *Salvia sahendica*. *Chemistry & Biodiversity*. 3 (8), pp: 917-922.
 20. Abu-Darwish, M.S., Cabral, C., Ferreira, I.V., Goncalves, M.J., Cavaleiro, C., Cruz, M.T., Albdour, T.H. and Salgueiro, L. (2013) Essential oil of common sage (*Salvia officinalis* L.) from Jordan: Assessment of safety in mammalian cells and its antifungal and anti-inflammatory potential. *Bio Med Research International*. 2013, 1-9. 538940. doi: 10.1155/2013/538940.
 21. Karcioğlu, L., Tanis, H., Comlekcioglu, N., Diraz, E., Kirecci, E. and Aygan, A. (2011) Antimicrobial activity of *Salvia trichoclada* in Southern Turkey. *International Journal of Agriculture & Biology*. 13(1), pp: 134-136.
 22. Paknejadi, M., Foroohi, F. and Yousefzadi, M. (2012) Antimicrobial activities of the essential oils of five *Salvia* species from Tehran province, Iran. *Journal of Paramedical Sciences*. 3(2), 12-18.
 23. Parameter, J.R.; Whitney, H.S. (1970). Taxonomy and nomenclature of imperfect state in *Rhizoctonia solani* biology and pathology. J.R. Parameter Ed. Univ. California Berkeley 7-10.
 24. Ellis, M. B. Dematiaceous Hyphomycetes. (1971) Commonwealth mycological Institute England. D. pp 608.
 25. Pitt, J. I. and Hocking, A. D. (1985). Fungi and food spoilage. Blackie Academic & professional: pp 593.
 26. DeHoog, G. S. and Guarro, J. (1995). Atlas of clinical fungi. Universitat Rovirai Virgili, Spain, p 720.
 27. Barnett, H. I.; Hunter, B. B. (2006). "Illustrated Genera of Imperfect Fungi" Burgess Publishing Company, 241 4th Edition, The American Phytopathological Society, St. Paul Minnesota.
 28. Rios, J. L.; Recio, M. C. and Villar, A. (1987). Antimicrobial activity of selected plants employed in the Spanish Mediterranean area. *J. of Ethnopharmacology*, 21 (2), pp: 139-152.
 29. Sousek, J.; Guedon, D.; Adam, T.; Bochorakova, H.; Taborska, E.; Valka, I. And Simanek, V. (1999). Alkaloids and organic acids content of eight fumaria species. *Phytochemical analysis*, 10 : 6-11.
 30. Gray, B. (1959). The relation of Fungi human affairs. Henry Holt and So. Inc. USA.
 31. Guneser, S.; Atici, A.; Koksal, S. and Yamam, A. (1994). Mold allergy in Adna, Turkey. *Allergy Immunol. Tath. Madril* 22(2), pp: 52-4.
 32. Parisa Badiiee, et al. (2012) Comparison of *Salvia officinalis* L. essential oil and antifungal agents against candida species *Journal of Pharmaceutical Technology & Drug Research* ISSN 2050-120X. doi: 10.7243/2050-120X-1-7.
 33. Croteau R (1986) Biochemistry of monoterpenes and sesquiterpenes of the essential oils. *Herbs, spices and medicinal plants: Recent advances in botany, horticulture, and pharmacology* 1: 81-135.
 34. Guenther E (1972) The production of essential oils: methods of distillation, effleurage, maceration, and extraction with volatile solvents. In: Guenther E (ed) *The essential oils. History, origin in plants, production analysis* 1: 85-188.
 35. Pinto, E.; Salgueiro, L. R.; Cavaleiro, C.; Almeida, A.; Goncalves, M. J. (2007). In vitro susceptibility of some species of yeast and filamentous fungi to essential oils of *Salvia officinalis*. *Industrial Crops and products*. 26(2), 135-141.
 36. Ileana, C. F.; Opre, E. (2006). Ethanol extracts of *Salvia officinalis* exhibit antifungal properties against *Saccharomyces cerevisiae* cells. *chimie. Anul. I.* 51-55.
 37. Al-hayally, (2011) Investigation of the genetic effect extracted from the sage plant (*Salvia officinalis*) *Journal of Rafidain Science*. 22(1), 127-141.