



Isolation And Identification Of Fungi Isolated From Tinea Infections On Head In Babylon City

Ruwaida A Al- Aqili¹ - Ebtesam Th. Jeaz²

Al-Qadisiyah University/College of Education/ Department of Biology.

Abstart

This study was conducted in the laboratory of the College of Education at Al-Qadisiyah University and the College of Engineering Technologies at Al-Qasim Green University for the period 11/3/2022 until 4/6/2023 with the aim of investigating the fungi that cause ringworm of the head (*Tinea capitis*). 125 samples were collected from patients visiting the dermatology consultation in Al-Hashimiya General Hospital and the private clinic of Dr. Najm Abdullah Al-Shammari / Babil Governorate / Al-Musayyib for age groups between (1-30) years and for both sexes, as the samples taken from the scalp included scales and hair, under the direct supervision of the specialist doctor, and the scale samples were taken by scraping, as they were sterilized. The affected area was treated with ethyl alcohol at a concentration of 70%, and then the crusts were scraped off from the edge of the focus affected by the fungi using a sharp, sterile blade. As for the hairs, they were taken using sterile forceps. The samples were diagnosed by direct microscopic examination using KOH at a concentration of 10%, and the remaining part of the pathological samples were planted on plates containing the medium of Subroid Dextrose Agar. Direct microscopic examination of the fungi showed positive results with an infection rate of 36% out of a total of 125 samples, while laboratory culture showed positive results with an infection rate of 48.8% out of a total of 125 samples. The results of phenotypic examinations of the isolated skin fungi showed that they belong to the two genera, namely *Trichophyton* and *Microsporum*. The results showed that the genus *Microsporum* was the most frequently diagnosed specimen, compared to the genus *Trichophyton*, the least of the patient specimens from which two species of *Microsporum* were isolated, namely *Microsporum. Canis* and *Microsporum. gypsum*, where *Microsporum.canis* had the highest percentage (44.44%) and the percentage of *Microsporum* fungi. *gypsum* (13.33%). As for the genus *Trichophyton*, four species were isolated from it: *Trichophyton.mentagrophytes*, *Trichophyton.tonsuranis*, and *Trichophyton. rubrum* and *Trichophyton .verrcosum*, with the highest percentage being 13.33% and *Trichophyton.mentagrophytes* being the lowest. *T.rubrum*(4.44%) .

Key words: Tinea capitis, Fungi .



عزل وتشخيص الفطريات المعزولة من سعفة الرأس في مدينة بابل

رويدة العكيلي

ابتسام الجعاز

جامعة القادسية/كلية التربية/قسم الأحياء

الملخص

أجريت هذه الدراسة في مختبر كلية التربية في جامعة القادسية وكلية التقانات الهندسية في جامعة القاسم الخضراء للمدة 2022/3/11 ولغاية 2023/6/4 بهدف التحري عن الفطريات التي تسبب سعفة الرأس (*Tinea capitis*). تم جمع 125 عينة من المرضى المراجعين لاستشارة الأمراض الجلدية في مستشفى الهاشمية العام والعيادة الخاصة للدكتور نجم عبدالله الشمري / محافظة بابل / المسيب للفئات العمرية ما بين (1-30) سنة ولكلا الجنسين، حيث شملت العينات المأخوذة من فروة الرأس القشور والشعر، وذلك تحت الإشراف المباشر للطبيب المختص، وتم أخذ عينات القشور عن طريق الكشط، كما تم تعقيمها. وتم علاج المنطقة المصابة بالكحول الإيثيلي بتركيز 70%، و تم كشط القشور من حافة البؤرة المتأثرة بالفطريات باستخدام شفرة حادة ومعقمة. أما الشعيرات فقد تم أخذها باستخدام ملقط معقم. تم تشخيص العينات عن طريق الفحص المجهرى المباشر باستخدام KOH بتركيز 10%، وتم زرع الجزء المتبقي من العينات المرضية على صفائح تحتوي على وسط Subroid Dextrose Agar. وأظهر الفحص المجهرى المباشر للفطريات نتائج إيجابية بنسبة إصابة 36% من إجمالي 125 عينة، بينما أظهرت الزراعة المخبرية نتائج إيجابية بنسبة إصابة 48.8% من إجمالي 125 عينة. وبينت نتائج الفحوصات المظهرية للفطريات الجلدية المعزولة أنها تعود إلى الجنسين وهما *Trichophyton* و *Microsporum* ظهرت نتائج أن الجنس *Microsporum* كانت أكثر العينات التي تم تشخيصها مقارنة بالجنس *Trichophyton* أقل العينات المرضى التي تم عزل نوعين من *Microsporum* هما *Microsporum. Canis* و *Microsporum. gypsum* حيث كان *Microsporum. canis* أعلى نسبة (44.44%) ونسبة الفطر *Microsporum. gypsum* (13.33%)، أما الجنس *Trichophyton* فقد عزل منه أربعة أنواع هما *Trichophyton. mentagrophytes* و *Trichophyton tonsurans*. و *Trichophyton. rubrum* و *Trichophyton. verrucosum*. وكانت *Trichophyton. mentagrophytes* أعلى نسبة (13.33%) وأقل نسبة *Trichophyton. rubrum* (4.44%).

الكلمات المفتاحية: سعفة الرأس، الفطريات

Introduction

Diseases caused by skin fungi are divided according to the site of infection, including: *Tinea capitis*, *Tinea corporis*, *Tinea manuum*, *Tinea facie*, *Tinea pedis*, and *Tinea cruris*. Therefore, skin fungi are the main cause of one of the most important skin diseases, the incidence of which has increased at a rate of significant over the past years (White, and Achterman, 2011). It is noteworthy that superficial fungal infections are common all over the world, with a higher incidence of infection in tropical and subtropical regions. This is due to providing conditions of environmental temperature and the presence of humidity, closed shoes, and tight clothing, which provide an opportunity for the infection to spread at a higher rate (Verma et al. ,202).

In geographical areas with high prevalence, the rate of infection with skin fungi can reach 20-25%, and various factors such as age, gender, social and



economic level, and cultural customs can contribute to the occurrence of infection (Chowdhardy et al., 2022).

The annual cost of treating these diseases was estimated at about \$500 million annually worldwide, and these diseases ranked second among diseases that affect the skin (El-Delasty et al., 2013).

Tinea capitis is a common infection that affects the hair and scalp and occurs mainly in children. Tinea capitis occurs due to either anthropophilic or zoophilic causes. Geophilic dermatophytes are less common throughout the world. *Trichophyton violaceum* and *Trichophyton tonsurans* are considered the most common types of fungi .

Dermatophiles are the most frequently isolated dermatophytes, while *Microsporum canis* is the most common dermatophyte fungus (Rodriguez et al., 2021).

The ability of pathogenic fungi to cause disease depends on several factors, including the surrounding environmental conditions, the host's immunity, the virulence of the pathogen, and the incidence of chronic diseases such as diabetes, lymphitis, and others. Virulence is defined as those means and mechanisms through which the fungus can cause infection and penetrate the host's body, and includes toxins, enzymes, the ability to adhere, and others. As these virulence factors increase the risk of the fungus towards humans and animals, especially the pathogen coming from a strange environment to another environment, which makes it dangerous and may become epidemic for the reasons mentioned above (Leitao, 2020).

Azoles and polyenes are known as chemical compounds that kill or inhibit fungi through their interference with the metabolism of the cell membrane, especially with Ergosterol, which causes distortion in the cell membrane of the fungal cell and causes it to lose its contents and then die. These antifungals were used with skin fungi and gave mixed results. Studies have indicated improvement in skin fungi against these antibiotics (Peyclit et al., 2021).

While other studies have indicated that dermatophytes are resistant to these antifungals, this is due to the ability of dermatophytes to produce enzymes that have the ability to transform the antibiotic into an inactive substance, and currently have the ability to destroy host cells (Arastehfar et al., 2020; Duxbury et al., 2020).

Materials and Methods

125 samples were collected from the private clinic of Dr. Najm Abdullah Al-Shammari in Al-Musayyib District / Babil Governorate, as well as from Al-Hashimiya General Hospital for people suffering from ringworm for a period



ranging from November 20/11/2022 until April 6/4/2023. The study included 67 samples. Of the males and 43 samples of females, for age groups ranging from one year to thirty years, by taking scales and hair from the affected area, a questionnaire form was recorded that included information about the infected person (gender, age, occupation, residence, accompanying symptoms).ollection of samples

Cultivation of samples

planted part of the fungal samples, represented by scales and hair, which were not treated with a potassium hydroxide solution, as they were grown on SDA medium, and then the antifungal Cycloheximide was added to it to prevent the growth of saprophytic fungi, and the antibiotic Chloramphenicol was also added to prevent bacterial growth. For the hair sample, two capillaries were taken or Three hairs were placed at equal distances in the dish, and then the dishes were kept in the incubator at a temperature of 28°C for 2-4 weeks to observe the appearance of fungal growth (Kannan et al., 2006).

.Phenotypic diagnosis of fungus

The fungal species were purified by the transfer of part of the developing colonies in the first form by a Loop To Petri dishes containing media (SDA) and incubate at 28°C for (7 days), repeating this process several times to obtain pure colonies (Kown-Chung & Bennett, 1992). The diagnosis was based on microscopic characteristics by preparing slides by moving a portion of the colony by a sterile loop into the slide and by adding a drop of blue dye. The samples were then examined under a microscope to examine the hyphae whether they are divided or not, the Conidia and the morphological characteristics of the colony such as shape, color and size, and the diagnosis was based on the following taxonomic Keys (Moustafa, 1982; Afzal *et al.*, 2013; Kidd *et al.*, 2016; Carmen & Sciortino, 2017).

Result and Discussion.

Total number of fungi distributed by location

ulture on SDA medium showed the presence of 64 positive samples and 61 samples. Negative by 51.2% and 48.8%, respectively. 125 pathological samples were collected from infections diagnosed clinically by specialist doctors for patients infected with skin fungi at Al-Hashimiya General Hospital and the private clinic of Dr. Najm Abdullah Al-Shammari for the period between 11/20/2022 to 4/6/2023 for age groups between (1-30). 1 year and for both sexes, and the results of direct microscopic examination using 10% KOH showed the presence of 45 positive skin samples, representing 64% of the total



number of samples, and 80 negative skin samples for examination, meaning 36%, while the results of laboratory c

| Test type | Positive samples | | Negative samples | | Total |
|--------------------------------|------------------|--------|------------------|-------|-------|
| Direct microscopic examination | 45 | %64 | 80 | %36 | %100 |
| Laboratory culture | 64 | %51.25 | 61 | %48.8 | %100 |

Table (11) shows that the highest percentage of those infected with ringworm of the head, whose ages ranged between (6-10) years, was 30% during the study, and these age groups showed that the number of males was higher than that of females infected, i.e. it reached 35% and 37.5%. respectively .

This percentage agreed with what was found by (Shorouk, 2013), as it found that the age group of 6-10 years was the most vulnerable to depression at a rate of 47.27%, while it did not agree with what was found by (Maha, 2021), as it showed that the age group (1-5) They are most susceptible to infection, at 44% .

Then comes the age group (1-5) with a rate of 28.125%, followed by the age group (11-15) with a rate of 18.75%, the age group (16-20) with a rate of 10.93%, and the age group (21-25) with a rate of 6.25%

Tabal -11- Percentages of age groups of people infected with skin fungi .

| Age group (the year) | Total | | Sex | | Percentage |
|-------------------------|-------|----------|------|----------|------------|
| | male | Feminine | male | Feminine | |
| 1-5 | 9 | 6 | 15 | 22.5 | 23.43 |
| 6-10 | 12 | 7 | 19 | 30 | 29.68 |
| 11-15 | 7 | 4 | 11 | 17.5 | 16.66 |
| 16-20 | 6 | 3 | 9 | 15 | 12.5 |
| 21-25 | 4 | 2 | 6 | 10 | 8.33 |
| 26-30 | 2 | 2 | 4 | 5 | 8.33 |
| total | 40 | 24 | 64 | 24 | 40 |



Picture No. (1) represents people infected with skin fungi by residence with tinea capitis. A- A 9-year-old female in (the rural area) infected with fungus *T. tonsorans* B. A 12-year-old boy infected with the *T. mentagrophytes* fungus in a rural area. C- A 30-year-old girl infected with the fungus. *M. canis* in the center of the province .

The reason for children's infection is due to their incomplete mental awareness, lack of interest in personal hygiene, poor health awareness, as well as the child's immune status. Also, direct contact between children contributes to transmitting the infection among them (Stevens, 1996). Its appearance in children and its infrequency in adults is due to the excessive formation of fatty acids in the scalp, which form an inhibitory compound for skin fungi. Therefore, the tendency to recover from this disease in adulthood is due to the change in the composition of fats in adults (Cremer, 1997).



3-2-4 Injury according to area of residence Home

The results of the study showed that the rate of infection with the fungi that cause tinea capitis differed according to the region of residence (rural, city) as in Table No. (4-4), where the highest rate of infection was in patients coming from rural areas at 62.5% (40) isolates, and the lowest rate was 37.5% (24 isolates). The infection rate in urban areas is 37.5% (24).

These results agreed with what was found by (Al-Zubaidi, 2019) and (Rashid, 2018), who found that infection in rural areas was higher than in urban areas, and did not agree with the findings of Al-Shaheen (2014) and Mustafa (2009). Al-Qaisi (2006) indicated that people who lived in rural areas were more vulnerable to infection than those who lived in city areas due to the economic and social level, the deteriorating health condition, and the lack of attention to the health condition, as well as the environmental conditions that are a major factor in causing diseases

Tabal -13- arelationship between infection with skin fungi and residence area (Countrysid ,City) .

| Residence area | Number of isoates | Percentage |
|----------------|-------------------|------------|
| Countryside | 40 | 62.5 |
| City | 24 | 37.5 |
| Total | 64 | 100 |

Distribution of isolates of dermatophytes causing tinea capitis

The results showed that the skin fungi that cause ringworm belong to the genera *Microsporum* and *Trichophyton*, and 26 isolates (57.77%) of *Microsporum* were the most common in causing infection, while the genus *Trichophyton* recorded 19 isolates, representing a rate of 42.22% .

This study agreed with what was found by Al-Jumaili (2008), who recorded that the genus *Microsporum* was more frequent than the genus *Trichophyton*, and it did not agree with Azheen(2022), Al-Qaisi (2006) and Maha (2021) who showed that the genus *Trichophyton* was more frequent than the genus *Microsporum*, and this The difference may be due to the size of the sample taken, the sample collection areas, and the time of sample collection .

The results in (Table 4-7) showed that the fungus *M. canis* is one of the most frequently isolated skin fungi, with approximately 20 isolates, or a percentage



of (44.44%), followed by the fungus *M.gypsum*, with 6 isolates, a percentage of (13.33%), while the fungi *T.tonsurans* and *T. mentagrophytes* recorded 6 isolates of each fungus, at a rate of (13.33%), then the fungus *T. verrucosum* was isolated at a rate of 5 isolates (11.11%), while the fungus *T. rubrum* was the least frequent species, at a rate of 2 isolates, at a rate of (4.44%) .

Table-15- Genera of dermatophytes .

| Fungi genera | Number | Percentage |
|------------------|--------|------------|
| Microsporum ssp | 26 | 57.77 |
| Trichophyton ssp | 19 | %42.22 |
| Total | 45 | %100 |

Schedule -16- Types of skin fungi isolated from patients.

| Mushroom type | Number | Percentage |
|------------------|--------|------------|
| M.canis | 20 | 44.44% |
| M.gypsum | 6 | 13.33% |
| T.mentogrophytes | 6 | 13.33% |
| T.tonsurans | 6 | 13.33% |
| T.verrcosum | 5 | 11.11% |
| T.rubrum | 2 | 4.44% |
| Total | 45 | 100% |

The results of the current study showed that the isolated dermatophyte species belong to both sexes, and this result is consistent with (Ahmed, 2022) and (Naik et al., 2019).

It has been shown from our current study that the fungus *M.canis* was one of the most isolated fungi with a rate of 44.44% (20 isolates) and causes tinea capitis. The fungus *M.canis* is the most frequently isolated species. This may be due to direct and indirect contact with pets such as cats. Dogs, livestock, etc., because it is a zoophilic fungus, causing many skin infections .

This result is consistent with (Shorouk, 2013), who found that the fungus *M.canis* is the most common at a rate of 49.09%, and the study of Kadian and



his group (2010) in Kuwait that the highest rate of infection with ringworm of the head is due to the fungus *M.canis*, so the infection rate with this fungus was 61.7. % for a study conducted on 903 children in 15 primary schools, as well as a study (Khattab et al., 2022) where they found that *M. canis* is the most common fungus at a rate of 38.5%.

References

1-Al- harbi ks , Joshi N, Singh Y , Kazmi Al, Abbasi FA, Alzare Sl, . Molecular exploration of hidden pleiotropic activities of azoles on dermatophytes in human tinea Corporis infection. Journal of Medical Mycology, **2022**, 101311.

Azheen, Berri Latif Muhammad (2022). Study of the phenotypic characteristics of the skin fungus that causes ringworm Tinea Corporis and the molecular diagnosis of the fungus Trichophyton mentagrophytes in the city of Kirkuk. Master's thesis - College of Science - Kirkuk University - 47 pages.

Ahmed, Hoda Abdel Rahim Abdel Karim (2022). Comparison of the inhibitory effectiveness of biosynthesized silver nanoparticles from plant extracts on the fungus T.mentagrophytes, Master's thesis, College of Veterinary Medicine, Tikrit University

2-Arastehfar, A., Lass-Flörl, C., Garcia-Rubio, R., Daneshnia, F., Ilkit, M., Boekhout, T., ... & Perlin, D. S. (2020). The quiet and underappreciated rise of drug-resistant invasive fungal pathogens. Journal of Fungi, 6(3),138

Al-Zubaidi, Alaa Abdel-Wadud Ahmed (2019). Isolation of active compounds from the fungus Ganoderma lucidum and identification of their inhibitory effectiveness against some skin fungi isolated from patients attending Salah al-Din General Hospital, Master's thesis. College of Science, Tikrit University

Al-Shahri, Nazim Allawi (2014). Characterization of extracts of some types of algae isolated from the water of the Tigris River and their effect on skin fungi of patients in Nineveh Governorate. Doctoral dissertation. College of Science. Tikrit University.

Afazl, H.; Shazad, S., and Nisa, S. Q. U. (2013). Morphological identification of Aspergillus species from the soil of Larkana District (Sindh, Pakistan). Asian Journal of Agriculture and Biology, 1(3): 105-117.

3-Achterman, R. R., & White, T. C. (2011). Dermatophyte virulence factors: identifying and analyzing genes that may contribute to chronic or acute skin infections. International journal of microbiology



Al-Jumaili, Afrah Qasim Saleh (2008). The effect of some plant extracts on the growth of fungi isolated from fungal infections in Tikrit General Hospital. Master Thesis. Faculty of Education. Tikrit University.

Al-Qaisi, A'raf Harith Abdel-Wahhab (2006). Epidemiological study and bacterial interaction of skin fungi Dermatophytes. Master Thesis. College of Science for Girls. Baghdad University.

4- Chowdhary A, Singh A, Kaux Ang, Khurana A. The emergence and World-Wide Spread of the Species *Trichophyton indetinea* causing difficult to treat dermatophytosis: A new challenge in the management of dermatophytosis. *Plos Pathogens*. **2022**;18(9):e1.10795

Carmen, V. and Sciortino, J.R. (2017). Atlas of clinically important fungi. Wiley Blackwell. USA.

5- Duxbury, S. J., Bates, S., Beardmore, R. E., & Gudelj, I. (2020). Evolution of drug-resistant and virulent small colonies in phenotypically diverse populations of the human fungal pathogen *Candida glabrata*. *Proceedings of the Royal Society B*, 287(1931), 20200761.

6- El-Dilasty, E. M. ;Ahmed, M. A. ;Okasha, N. ;Mansour, S. F. ;El-Dek, S ;Abd El-Khalek, H. and Youssif, M. H. (2013). Antifungal activity of zinc oxide nanoparticles against dermatophytic lesions of cattle. *Romanian J. Biophys.* 23(3): 191-202-

7-Khattab S, Abd El-Fadeal N, Azab H. Molecular identification of *Microsporum canis* isolates and detection of subtilisin genes from dermatophytosis. *Microbes Infect Dis* **2022**; 3(4): 1067-1073

8-Kidd, S., Halliday, C., Alexiou, H. and Ellis, D. (2016). Descriptions of medical fungi /. Edition: Third edition. Page: 126.

Kadian ,R.,El- Gamal , A.H.,Al-Mehnna ,B.,Al- Otaibi ,S., Najem , N., (2010). Tinea Capitis in Al-Ahmadi Province of Kuwait : ASurvey of Elmentary School children .*Iranian journal of dermatology* .Vol.13,No.1,6-8 .

Kannan, P. ;Janaki, C. and Selvi, G. S. (2006). Prevalence of dermatophytes and other fungal agents isolated from clinical samples. *Indian J.. Med.Microbio.*,24(3): 2012 – 2015 .

9-Kown- Chung, K. J. and Bennett, J. E. (1992). Medical Mycology. Lea. and Febiqer. 745Pp.MoubPress .



10- Leitão, J. H. (2020). Microbial virulence factors-11Dermatophytes Species Isolated From Clinical Specimens. *Journal of Inflammatory Diseases*. 2022; 26(1):35-42.

Maha, G., Hawazin ,A. (2021) . Assessment of antifungal activity of AgNPs ,Apple cider vinegar and Grisofulvin in Tinea capitis infections of children on some immunological and hematological paramenters in vitro and vivo .Athesis – College of science - Tikrit University

Mustafa, Berri Abdel Latif (2009). An epidemiological and diagnostic study of skin fungi in the city of Kirkuk and the effect of onion plant extracts on the isolated fungi. Master's thesis, College of Science, Tikrit University, Republic of Iraq.

Moustafa, A.F.(1982). Taxonomic studies on the fungi of Kuwait. *Aspergilli*. *Kuw. J. Sci*, 9: 245-260 .

11- Naik, P.S, Mangala, G.K., & Lava, R. (2019). Spectrum of Dermatophytic Fungal Infection in Tertiary Care Hospital,Davanagere. *Int. J.Curr.Microbiol.App.Sci* 8(3): 165-171

12-Peyclit, L., Yousfi, H., Rolain, J. M., & Bittar, F. (2021). Drug Repurposing in Medical Mycology: Identification of Compounds as Potential Antifungals to Overcome the Emergence of Multidrug-Resistant Fungi. *Pharmaceuticals*, 14(5), 488.

13- Rodríguez-Cerdeira, C.; Martínez-Herrera, E.; Szepietowski, J.C.; Pinto-Almazán, R.; Frías-De-León, M.G.; Espinosa-Hernández,

V.M.; Chávez-Gutiérrez, E.; García-Salazar, E.; Vega-Sánchez, D.C.; Arenas, R.; A Systematic Review of Worldwide Data on Tinea Capitis: Analysis of the Last 20 Years. *J. Eur. Acad. Dermatol. Venereol*. **2021**, 35, 844–883

Rasheed, Sarah Omran (2018). Evaluating the therapeutic efficacy of some plant extracts on clinical isolates of skin fungi, while studying virulence factors. Doctoral dissertation. Department of Life Sciences, College of Science. Tikrit University.

Stevens, D.L. (1996). Cecil Textbook of Company Medicine. 20th ed., Vol. 2. Tokyo. W.B. Sanders.

14- Ullah, Z.; Gul, F.; Iqbal, J.; Abbasi, B.A.; Kanwal, S.; Chalgham, W.; El-Sheikh, M.A.; Diltemiz, S.A.; Mahmood, T. Biogenic Synthesis of Multifunctional Silver Oxide Nanoparticles (Ag₂ONPs) Using *Parietaria alsinaefolia* Delile Aqueous Extract and Assessment of Their Diverse Biological Applications. *Microorganisms* **2023**, 11, 1069



15-Verma, S.B.; Panda, S.; Nenoff, P.; Singal, A.; Rudramuruthy, S.M.; Uhrlass, S.; Das, S.; Bisherwal, K.; Shaw, D.; Vasani, R. Theunprecedented epidemic-like scenario of dermatophytosis in India: I. Epidemiology, risk factors and clinical features. Indian J. Dermatol. Venereol. Leprol. **2021**, 87, 154–175