Identification of Causative Fungal Species of Tinea Capitis among Primary School Children in Qalubia Government

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Abstract

Background: Tinea capitis (T.capitis) is a disease caused by superficial fungal infection of the scalp, eyebrows and eyelashes, with a propensity for attacking hair shafts and follicles. There is certainly ample evidence that there is massive contamination of school rooms by viable spores in classes where cases have occurred.

Objective: Determine clinical and mycological profile of T.capitis among primary school children and evaluate the sensitivity of KOH microscopy and culture.

Patients and Methods: A total of one hundred children were examined for evidence of T.capitis in Dermatology Outpatient Clinic of Benha University Hospital. They were exposed to KOH examination and cultures on Sabouraud’s dextrose agar (SDA) and Dermasel agar.

Results: The results from microscopic examination by KOH were 64%, from culture by SDA were 80% and culture on Dermasel agar were 82% positive. Trichophyton mentagrophytes was isolated in 25.7%, M.canis in 24.4%, T.violaceum in 14.6%, both M.audouinii and E. floccosum in 9.7%, both T.schoenleinii and T.tonsurans in 5.6.1% and lastly T.rubrum in 33.7%.

Conclusion: Dermatophytes cultured on Oxoid Dermasel agar showed characteristic colonial morphology with typical pigmentation, less chance for contamination and rapid diagnosis. T.mentagrophytes (zoophilic) was the most common isolated dermatophytes (25.7%), followed by M.canis (zoophilic) (24.4%).

Key Words: Tinea capitis – KOH – SDA – Dermasel agar.

Introduction

T.CAPITIS is an infection caused by dermatophyte fungi (usually species in the genera Microsporum and Trichophyton) of scalp hair follicles and the surrounding skin [1]. T.capitis is a highly contagious disease widely distributed throughout the world, but more prevalent in hot humid tropical climates than in cold dry regions. The incidence of the disease is influenced by various factors such as race, socioeconomic conditions, cultural patterns and public health measures [2,3]. Sources of infection and mode of transmission can occur through use of contaminated barbershop instruments, combs, hair brushes and hats and also by direct contact with contaminated theater seats or other furniture [4].

Microscopy examination and/or fungal culture should be used to confirm the clinical diagnosis of T.capitis because of the extended nature of most treatment regimens [5].

Microscopy yields rapid results; however, this method cannot discriminate between different types of dermatophytes, demands some basic skills in execution and analysis of the specimen. Furthermore, it may give rise to false-negative findings in early or inflammatory lesions [6].

The definitive identification of the causative dermatophyte in T.capitis requires isolation of the fungus by culture. Several media are available, including Sabouraud’s dextrose agar with chloramphenicol (to inhibit bacterial growth) and cycloheximide (to inhibit nonpathogenic fungi growth). Sabouraud’s dextrose agar is an acidic pH medium for the isolation of dermatophytes, other fungi and yeasts [7]. Dehydrated culture media Dermasel agar base and supplement, which is a selective medium for dermatophyte fungi recommended for the examination of hair, skin scrapings, nails, etc [8].

Aim of work:

The aim of this study was to determine clinical presentation, age and sex distribution of T.capitis among school going children, and to identify the different causative fungal species and compare the results of microscopic examination, conventional cultures (Sabouraud’s dextrose agar); culture on specific media (dermasel agar and supplement) for rapid and accurate diagnosis.

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Patients and Methods

This cross-sectional study was carried out on primary school children from January 2012 to December 2012. A total of one hundred clinically suspicious cases were selected from Dermatology Outpatient Clinic of Benha University.

All patients were subjected to the following:
1- History taking including (age, sex, residence and contact with animals). 2- Dermatological examination (skin, hair, nails and mucous membrane) to detect any associated dermatological diseases. Inclusion criteria: Children were included if their age ranged from 4-12 years and had sign of T.capitis (alopecia, scaling, purities). Exclusion criteria: Children had known dermatological disease such as psoriasis or eczema or had been treated for T.capitis within the previous one month. An informed consent, which was approved by Ethics Committee Human Research of Benha University, was signed by a parent of each patient.

Specimen collection:

The specimen was collected after cleaning the scalp with 70% ethyl alcohol to reduce the likelihood of bacterial contamination and improve the chance for fungal detection. Hair was then epilated by using forceps and was collected from the active edge of the lesion and from the centre as well. Dull, grey or broken hairs were selected and hair stumps were pulled. In cases of scaly type, scales were scraped from the surface by the edge of a clean glass slide. In cases of kerion, the lesion was washed with saline solution instead of alcohol to avoid pain and irritation. Wet sponge was used for cleaning as cotton dressings might produce fibers that made microscopic examination difficult. In case of favus, thick crusts were scraped from the surface of the scalp gently by a clean glass slide. Cutting of hair was avoided as the infection is usually confined to the root and very near to the scalp surface. Hair and scales were then collected in sterile dry containers then transported to the laboratory for mycological examination.

All samples were subjected to the following:
1- Potassium hydroxide preparation, by emulsifying the specimen in a drop of 15% KOH on a microscope slide. The purpose of KOH was to clear out any background scales or cell membranes that may be confused with hyphal elements. Clearing was accelerated by heating the mixture gently over the flame of a Bunsen burner. A cover slip was applied and the specimen was examined for the presence of narrow, regular hyphae that characteristically break up into arthroconidia. Mosaic arrangement of spores was seen on the surface of the shaft (ectothrix infection) or hyphal fragments and arthroconidia was seen internally (endothrix infection).

2- Culture on Sabouraud’s dextrose agar (Oxoid). The cultures were incubated at room temperature for 4-6 weeks and observed regularly for growth. The fungal isolates were identified on the basis of duration of growth, surface morphology, pigment production on the reverse, microscopic examination in lacto phenol cotton blue preparation and slide culture; urease and hair penetration test whenever necessary.

3- Culture on Dermasel agar base and suppleme T (Oxoid). The medium was incubated at 22-30°C and examined at regular intervals for 2-4 weeks.

Statistical analysis:

The clinical and laboratory data were tabulated, coded then analyzed by using Microsoft Office Excel 2010 software program for figures and IBM SPSS (Program statistical package for social science) version 19 software (SPSS Inc., an IBM Company Headquarters, 233 S. Wacker Drive, 11 th floor, Chicago, Illinois 60606).

Results

Descriptive results:

In this study 100 children were clinically suspected for T.capitis, 62 males and 38 females, their age ranged from 4-11 years with the mean age of 6.4±2.2 years, 82 children were positive by culture on Dermasel agar and 18 children were negative, 72 lived in rural areas and 10 lived in urban areas. It is more common among the children aged 4-6 years 42.7%. The males were more affected than females. Males were more affected in rural and urban areas 62.5% and 70% respectively than females 37.5% and 30% (Table 1). Scaly T.capitis was the commonest clinical type, it account for (47.6%), black dot type in (40.2%), favus and kerion in (6.1 %) for each.

Laboratory results:

Dermasel agar culture was able to detect 82 out of 100 of clinically suspicious cases of T.capitis (82%) followed by culture on SDA (80%) and lastly came direct KOH microscopic examination which detect only (64%) of cases but this was insignificant (Table 2).

Different isolated dermatophytes causing T.capitis by culture on Dermasel agar and culture on SDA. T.mentagrophytes (zoophilic) was the most common isolated dermatophytes (25.7%),
followed by M. canis (zoophilic) (24.4%), T. violaceum (14.6%), both M. audoniuii and E. floccosum (9.7%) for each, T. tonsurans and T. schoenlenii (6.1%) for each and lastly T. rubrum (3.7%) (Table 3). The most common isolated dermatophyte from rural areas was T. mentagrophytes (26.4%) and from urban areas was both T. mentagrophytes and M. canis (20%) for each (Table 4) (Figs. 1-7).

Fig. (1): Colony of microsporum canis on dermasel agar, the colony tend to be white and fluffy.

Fig. (2): The left side represent smooth and white variant of T. mentagrophytes culture on dermasel agar, there was no evidence of background pigmentation. The right side represents the colony of T. rubrum on Dermasel agar demonstrating a reddish pigmentation of the background agar.

Fig. (3): The left side represents the reverse of colony of T. mentagrophytes growing on dermasel agar. The right side represents the reverse of colony of T. rubrum. The comparative growth illustrating the distinct difference in pigmentation between the two species.

Fig. (4): E. floccosum microscopic examination showing, cellular macroconidia (2-5) produced singly or in small clusters.

Fig. (5): Microsporum canis microscopic examination showing macroconidia, macroconidia are spindle shaped and multicellular and characterized by a thick outer wall with rough surface and sharpened tips.

Fig. (6): Trichophyton mentagrophytes microscopic examination showing microconidia, they are many and arranged in loose clusters, macroconidia are present in small numbers, thin and pencil shaped.

Fig. (7): Trichophyton violaceum microscopic examination showing sterile hyphae.
Identification of Causative Fungal Species of Tinea Capitis

Table (1): Number of tinea capitis positive cases in relation to sex and residence.

<table>
<thead>
<tr>
<th>Sex</th>
<th>Male</th>
<th>Female</th>
<th>Total</th>
<th>Z</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Residence</td>
<td>No.</td>
<td>%</td>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>Rural</td>
<td>45</td>
<td>62.5%</td>
<td>27</td>
<td>37.5%</td>
<td>72</td>
</tr>
<tr>
<td>Urban</td>
<td>7</td>
<td>70%</td>
<td>30</td>
<td>30%</td>
<td>10</td>
</tr>
<tr>
<td>Total</td>
<td>52</td>
<td>63.4%</td>
<td>30</td>
<td>36.6%</td>
<td>82</td>
</tr>
</tbody>
</table>

Table (2): Results of different methods for diagnosis of tinea capitis.

<table>
<thead>
<tr>
<th>Methods of diagnosis</th>
<th>No.</th>
<th>%</th>
<th>Z</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microscopic examination by KOH</td>
<td>64</td>
<td>64.0%</td>
<td>Z1=1.33</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>+ve results by culture on Sabouraud’s dextrose agar</td>
<td>80</td>
<td>80.0%</td>
<td>Z2=1.44</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>+ve results by culture on Dermasel agar</td>
<td>82</td>
<td>82.0%</td>
<td>Z3=0.16</td>
<td>&gt;0.05</td>
</tr>
</tbody>
</table>

Table (3): Isolated dermatophytes causing tinea capitis by culture on SDA and Dermasel agar.

<table>
<thead>
<tr>
<th>Etiological agent</th>
<th>Sabouraud’s Agar</th>
<th>Dermasel Agar</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>T.mentagophytes</td>
<td>21</td>
<td>26.3%</td>
</tr>
<tr>
<td>M.canis</td>
<td>19</td>
<td>23.7%</td>
</tr>
<tr>
<td>T.violaceum</td>
<td>12</td>
<td>15%</td>
</tr>
<tr>
<td>M.audonii</td>
<td>8</td>
<td>10%</td>
</tr>
<tr>
<td>E.floccosum</td>
<td>8</td>
<td>10%</td>
</tr>
<tr>
<td>T.schoenleinii</td>
<td>5</td>
<td>6.3%</td>
</tr>
<tr>
<td>T.tonsurans</td>
<td>4</td>
<td>5%</td>
</tr>
<tr>
<td>T.rubrum</td>
<td>3</td>
<td>3.7%</td>
</tr>
<tr>
<td>Total</td>
<td>80</td>
<td>100.0%</td>
</tr>
</tbody>
</table>

Table (4): Different isolated dermatophytes causing tinea capitis in relation to residence.

<table>
<thead>
<tr>
<th>Etiological Agent</th>
<th>Isolated rural</th>
<th>Isolated urban</th>
<th>Total</th>
<th>Z</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>T.mentagophytes</td>
<td>19</td>
<td>26.4%</td>
<td>2</td>
<td>20%</td>
<td>21</td>
</tr>
<tr>
<td>M.canis</td>
<td>18</td>
<td>25%</td>
<td>2</td>
<td>20%</td>
<td>20</td>
</tr>
<tr>
<td>T.violaceum</td>
<td>11</td>
<td>15.3%</td>
<td>1</td>
<td>10%</td>
<td>12</td>
</tr>
<tr>
<td>M.audonii</td>
<td>7</td>
<td>9.7%</td>
<td>1</td>
<td>10%</td>
<td>8</td>
</tr>
<tr>
<td>E.floccosum</td>
<td>7</td>
<td>9.7%</td>
<td>1</td>
<td>10%</td>
<td>8</td>
</tr>
<tr>
<td>T.tonsurans</td>
<td>4</td>
<td>5.6%</td>
<td>1</td>
<td>10%</td>
<td>5</td>
</tr>
<tr>
<td>T.schoenleinii</td>
<td>4</td>
<td>5.6%</td>
<td>1</td>
<td>10%</td>
<td>5</td>
</tr>
<tr>
<td>T.rubrum</td>
<td>2</td>
<td>2.7%</td>
<td>1</td>
<td>10%</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>100%</td>
<td>10</td>
<td>100%</td>
<td>82</td>
</tr>
</tbody>
</table>

Discussion

T. capitis concerns infection of the scalp by dermatophytes of the genera Microsporum and Trichophyton. The causal agent cannot be determined in all children by means of clinical features alone [9].

In this study, males were more affected with T. capitis than females in rural and urban area, this study go in agreement with the study of Del Boz et al., [10] who reported that the majority of cases (61.5%) are boys. A possible explanation for this apparent predominance of cases in boys may be attributed to the fact that, male children have more contact with animals, and in some countries, rural boys help out on farms. Also, the ringworm is more visible in boys with short hair allowing easy access for circulating spores, and in Islamic countries, girls are infected less possibly because when they get older they keep their heads covered, in accordance with Islamic law [11].

The present work reflected that people in rural areas had a higher risk of infection with T. capitis than that lived in urban area, where 87.8% of patients lived in rural areas, and 12.2% lived in urban areas, this was significant. These results were in agreement with other authors [12-15] they reported that tinea capitis occurs predominantly in rural or suburban areas, and some of the factors associated with this increased frequency include, low standards of living, overcrowding, poor hygiene and low level of parents education are factors which increase susceptibility to T. capitis infection in rural areas [11].

In this study scaly type was the most frequent type in positive cases, this goes in agreement with another study [16], which mentioned scaly tinea capitis as the most common presentation, while other investigators [17,18], reported black dot to be the most frequent type. This difference may be attributed to the different types of isolated organisms and different geographical location.

In the present study, five cases of favus were recorded. This goes in agreement with other studies in different parts of the world which reported marked decrease or nearly complete absence of favus cases due to disappearance of Trichophyton schoenleinii due to improved socioeconomic conditions [19]. Gargoom et al., [16], reported that there has been a dramatic decrease in the incidence of favus with complete disappearance of T.schoenleinii and T.verrucosum as causative agents of T.capitis.

In this study the correlation between a positive microscopic examination of hairs and scales by KOH 64%, detection of species by primary culture on SDA was 80% and 82% were positive by culture on Dermasel agar. Garcia et al., [20], reported that the result from culture was more accurate than microscopic examination by KOH.
In our study culture on Dermasel agar was more specific with better topography, texture, color, and colony surface and reverse, less growth of contaminant bacteria and saprophytic fungi. Seyfarth et al., [22] suggested that Dermasel media for growth of dermatophytes have a pH of 6.8-7.0 rather than pH 5.6 as is often recommended in other media. This pH is better for the growth of some fungi and suppresses bacterial contaminants.

Dermatophytes cultured on Dermasel agar show characteristic colonial morphology with typical pigmentation, macroconidia and microconidia are typical for the species when studied microscopically [22].

In this study, T. mentagrophytes followed by M. canis were the most common isolated organisms, these results goes in agreement with other studies [10,19]. This may be attributed to predominance of T. mentagrophytes and M. canis was probably related to increase in the number of pets (particularly cats and dogs) and peoples working in farm [10].

In the current study zoophilic fungi represent 50% of total isolated dermatophytes (T. mentagrophytes 51.2% and M. canis 48.8%). This study goes in agreement with study of Emele and Oyeka [23] who reported that, the low level of enlightenment of these purely rural agricultural communities, coupled with associated crowded and unhygienic rural lifestyles and low economic conditions, might have enhanced the spread of the disease among the population located in these regions and the most frequent isolate was T. mentagrophytes 44.6% followed by M. canis 32.3% and T. verrucosum 23.1%.

Conclusion:

Tinea capitis affects males more than females, common among the children aged 4-6 years, and common among rural school children than urban ones. Dermatophytes cultured on Oxoid Dermasel agar showed characteristic colonial morphology with typical pigmentation, less chance for contamination and rapid diagnosis. T. mentagrophytes (zoophilic) was the most common isolated dermatophytes, followed by M. canis (zoophilic). Periodic surveillance of Tinea capitis among schoolchildren might be important to control possible outbreaks, especially in crowded areas.

References


