

## A Study of Fungal Isolates from Superficial Mycoses Cases in Pediatric Patients, Lucknow

Alok Kumar<sup>1</sup>, Ekta Rani<sup>2</sup>, Sanjeev Sahai<sup>3</sup>

<sup>1,2</sup> Department of Microbiology, MSY Medical College & Hospital, Meerut, India

<sup>3</sup> Department of Microbiology, Mayo Institute of Medical Sciences, Barabanki, India

**Abstract:** This study was an attempt to estimate the prevalence of fungal isolates in superficial mycoses cases in pediatric patients attending OPD of IIMS&R, Lucknow Uttar Pradesh. A prospective study over a period of six months was conducted from January 2017 to June 2017. The suspected cases of superficial mycoses were subjected to mycological examination with direct microscopy using 10% - 40% KOH depending on the types of samples (skin, nail, hair) processed and culture on Sabouraud's dextrose agar with chloramphenicol and cycloheximide (SDCCA) and also on Potato dextrose agar (PDA) medium. Causative agents were identified macroscopically and microscopically from the growth obtained on SDCCA and PDA. Direct microscopy revealed fungal elements in 93 (96.9%) cases whereas 81 (84.4%) were positive on culture. *Tinea corporis* 54 (56.2%) was the most common clinical types and male to female ratio in relation to clinical types was 2.6:1. Commonest age group affected were 11-14 years with 38 (34.6%) cases. Males were predominantly affected 38 (39.6%) and male to female ratio being 3.1:1. *Trichophyton mentagrophytes* 25 (30.9%) was the predominant isolate followed by *T. tonsurans* 17 (21%), *T. rubrum* 7 (8.6%), *M. audouinii* 5 (6.2%), *T. schoenleinii* and *M. ferrugineum* 3 (3.7%), *T. verrucosum* 2 (2.5%) and *T. interdigitale* 1 (1.2%) with Non Dermatophytes *Candida* spp. 3 (3.7%), *F. oxysporum* 1 (1.2%) no *Epidermophyton* species. A non-pigmented variant of *T. rubrum* was identified in this study. Both SDCCA and PDA were found equally effective in isolating fungal isolates from clinical samples in our study. We are reporting change in frequency of dermatophytes isolated from superficial mycoses cases in pediatric patients our region.

**Keywords:** Superficial mycoses, Non-pigmented variants, KOH mount

Date of Submission: 08-04-2020

Date of Acceptance: 23-04-2020

### I. Introduction

Infections caused by the pathogenic fungi invading the stratum corneum of skin, hair, or nails are called superficial fungal infections<sup>1</sup>. They are the most common encountered infections in the dermatology outpatient department. Among all the superficial fungal infections, dermatophytic infections have the highest prevalence in the developing countries with significant associated morbidity. Dermatophytic infections are caused by the *Trichophyton*, *Epidermophyton*, and *Microsporum* species. Based on the different body sites involved, the nomenclature varies as tinea corporis [Figure 1], tinea faciei [Figure 2], tinea cruris, tinea pedis, tinea manuum, tinea capitis with Endothrix [Figure 3], and tinea unguium [Figure 4]. These infections are predominantly seen in hot tropical countries like India. Various climatic factors and socioeconomic and host factors attribute to the increasing incidence of fungal infections among the pediatric population in recent years.



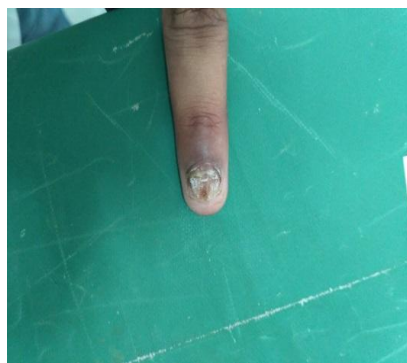
Figure 1: fungal infection of *T. corporis*



Figure 2: Tinea faciei



**Figure 3:** Tinea capitis and Endothrix in KOH mount



**Figure 4:** Tinea unguium

Environmental or climatic factors such as the El Nino phenomenon, increased duration of the summer season, increasing humidity, and geographical locations are responsible for the rising incidence of fungal infections. A myriad of host factors contributing to this rising trend are age, race, decreased rate of sebum production, immune status, any disruption in skin barrier, and associated atopic dermatitis<sup>2</sup>. Low socioeconomic status, poor hygiene, overcrowding, improper sanitation, lack of health education and awareness, and poor health-care facilities are the most important predisposing parameters<sup>3</sup>.

The infections are highly contagious in nature with a high rate of skin-to-skin and fomite transmission and recurrences. The increasing prevalence of the superficial fungal infections in the pediatric age group is surely a cause of concern because of its public health importance.

## **II. Materials And Methods**

This is a six month (January 2017 to June 2017) prospective study, was conducted at Integral Institute of Medical Science and Research, Lucknow, Uttar Pradesh. The study population comprised of 96 clinically suspected cases of superficial mycoses attending Dermatology OPD. All the clinically suspected cases of superficial mycoses referred to Department of Microbiology for isolation and identification of etiological agent were included in the study. Demographic details of every case and detailed history of onset of disease, duration of symptoms, trauma, patient's occupation, drugs, associated co-morbid conditions, family and personal history was taken. Enquiries were also made as to exposure to animals, cases or any other suspected sources.

## **III. Specimen Collection and Procedure**

The affected areas were swabbed with 70% alcohol. Skin scrapings or nail clippings or plucked hair was collected in clean white paper packets.

### **Skin scraping:**

Skin scrapings were collected by scraping across the inflammatory margin of the lesion including the healthy skin using sterile scalpel or clean slide. If vesicles are present, the top was removed with fine scissors and stored for further examination.

### **Nail scraping:**

Nail specimen was collected by taking the infected nail clippings and was scraped deeply enough to obtain the recently invaded nail tissue. In cases of paronychia i.e. where a yeast infection is suspected, exudate was expressed from the Paronychia folds by probing with a flat excavator and collecting on a swab previously moistened with sterile saline.

### **Hair plucking:**

Hair specimen was collected by plucking the infected hair including the base of hair shaft. The species most frequently associated with scalp ringworm cause the affected hairs to fluoresce under a wood's lamp and this is a useful means of selecting material.

### **Direct microscopic examination**

**KOH mount:** 10% KOH solution was used for skin and hair samples. 40% KOH for nail specimens and incubated overnight at 37°C for clearing. Clearing can be hastened by gently heating. As soon as the specimen has cleared, examined under microscope using the 10x and 40x objectives, for the presence of filamentous, septate, branching hyphae with or without arthrospores. In case of hair, type and arrangement of spores were

noted (ectothrix/ endothrix). *Tinea versicolor* infections were diagnosed by the presence of round yeast cells with short, stout and curved hyphae (spaghetti and meat ball appearance).

**Fungal culture**

All the samples were collected and inoculated on two sets of test tubes containing Sabouraud’s dextrose agar with chloramphenicol and cycloheximide and Potato dextrose agar. For *Tinea versicolor* infections SDA with sterile olive oil overlay was used. The fungal cultures have been identified by colony morphology, rate of growth and pigment production. Lactophenol cotton blue mount was done from the small bit of colony taken on clean glass slide and teased out using two teasing needles, to detect the presence of macroconidia, microconidia, chlamydospore and special hyphal structures. Confirmatory identification of the species was done by slide culture technique and Biochemical tests i.e. urease test, hair perforation test and rice grain test. Speciation of yeastlike fungi was done by gram’s stain, germ tube test, sugar fermentation and assimilation tests.

**IV. Observations and Results**

A total of 96 pediatric patients diagnosed as suspected case of superficial mycoses were enrolled in the study, none of them had any systemic disease. Most common clinical presentation was *T. corporis* 54 (56.2%) followed by *T. cruris* 13 (13.5%), *T. capitis* 10 (10.4%), *T. pedis* 06 (6.3%), *T. unguium* 5 (5.2%), *T. faciei* and *T. manuum* 04(4.2%) each. (Table 1.1)

**Table 1.1: Distribution of clinical types of superficial mycoses cases**

Clinical types	Total N (%)
<i>Tinea corporis</i>	54 (56.2)
<i>Tinea cruris</i>	13 (13.5)
<i>Tinea capitis</i>	10 (10.4)
<i>Tinea pedis</i>	6 (6.3)
<i>Tinea unguium</i>	5 (5.2)
<i>Tinea faciei</i>	4 (4.2)
<i>Tinea manuum</i>	4 (4.2)
<b>TOTAL</b>	<b>96 (100)</b>

On studying gender distribution of clinical types out of 96 cases, 63 (65.6%) were males while 33 (34.4%) were females with the male female ratio of 2:1, this difference was found to be statistically significant ( $p < .0001$ ). Among 63 (65.6%) male patients, 39 (40.6%) reported with *T. corporis* as the predominant clinical type, followed by *T. cruris* 10 (10.4%), *T. capitis* 05 (5.2%), *T. manuum* 04 (4.2%), *T. pedis* 03 (3.1%), and *T. unguium* 02 (2.1%). No case of *T. faciei* was reported from male patients.

Similarly in 33(34.4%) female patients, *T. corporis* was the predominant clinical type 15 (15.6%) followed by *T. capitis* 05 (5.2%), *T. faciei* 04(4.2%), *T. cruris* 03 (3.1%), *T. pedis* 03(3.1%), *T. unguium* 03(3.1%) and no case of *T. barbae*, *T. manuum* were reported from them. (Table 1.2)

**Table 1.2: Gender distribution of clinical types**

Clinical types	No. of cases N (%)	Male N (%)	Female N (%)	M:F ratio	P value
<b>T. corporis</b>	54(56.2)	39(40.6)	15(15.6)	2.6:1	0.18
<b>T. cruris</b>	13(13.5)	10(10.4)	03(3.1)	3.3:1	0.54
<b>T. capitis</b>	10(10.4)	05(5.2)	05(5.2)	1:1	0.45
<b>T. pedis</b>	06(6.3)	03(3.1)	03(3.1)	1:1	0.69
<b>T. unguium</b>	05(5.2)	02(2.1)	03(3.1)	1:1.5	0.44
<b>T. manuum</b>	04(4.2)	04(4.2)	-	N.A.	N.A
<b>T. faciei</b>	04(4.2)	-	04(4.2)	N.A.	N.A
<b>TOTAL</b>	<b>96(100)</b>	<b>63(65.6)</b>	<b>33(34.4)</b>	<b>2:1</b>	<b>&lt;.0001*</b>

N.A- not applicable due to low sample size, (\* $p \leq 0.05$ =significant)

On analysing distribution of clinical presentation among different age groups, out of total 96 cases, all types of tinea except *T. capitis* and *T. faciei*, were most common in 11-14 years age group with predominance of *T. corporis* 38 (39.6%) , followed by *T. cruris* 11 (11.5%), *T. pedis* 5 (5.2%), *T. manuum* 4 (4.2%), *T. unguium* 3(3.1%),

*T. capitis* 5 (5.2%) was the most common presentation in the age group of 6-10 years followed by 02 (2.1%) each in 1-5 and 11-14 years, and only 1(1.04%) case in <1 year age group. All cases of *T. manuum* were found in 11-14 years age group, however, no case of *T. cruris* were found in 6-10 years age group, *T. faciei* in < 1 year and *T. pedis* in < 1 year and 1-5 years age groups. (Table 1.3)

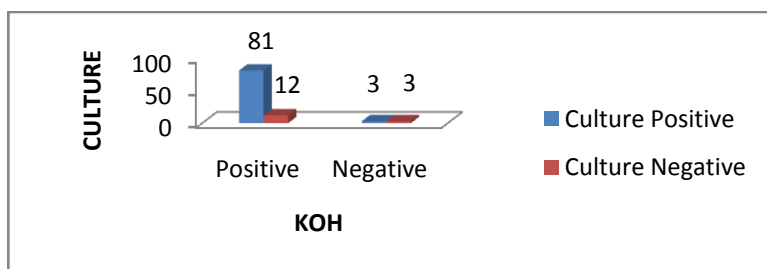
**Table 1.3: Age distribution of clinical types**

Clinical types	Age groups (in years)				
	Total N (%)	< 1 year N (%)	1-5 year N (%)	6-10 year N (%)	11-14 year N (%)
<b>T. corporis</b>	54(56.2)	03(3.1)	10(10.4)	03(3.1)	38(39.6)
<b>T. cruris</b>	13(13.4)	01(1.0)	01(1.0)	-	11(11.5)
<b>T. capitis</b>	10(10.4)	01(1.0)	02(2.1)	05(5.2)	02(2.1)
<b>T. pedis</b>	06(6.2)	-	-	01(1.0)	05(5.2)
<b>T. unguium</b>	05(5.2)	-	01(1.0)	01(1.0)	03(3.1)
<b>T. manuum</b>	04(4.2)	-	-	-	04(4.2)
<b>T. faciei</b>	04(4.2)	-	01(1.0)	01(1.0)	02(2.1)
<b>TOTAL</b>	<b>96(100)</b>	<b>05(5.2)</b>	<b>15(15.6)</b>	<b>11(11.4)</b>	<b>65(67.7)</b>

On direct microscopic examination by KOH mount, out of the total 96 samples, 93 (96.9%) samples were positive, while only 3 (3.1%) were negative, and 81 (84.4%) were culture positive and 12 (12.5%) were negative. All 3 KOH negative samples turned out to be culture negative too. (Table & Figure 1.4)

**Table 1.4: KOH and Culture results**

KOH Mount	Total N (%)	Culture	
		Positive N (%)	Negative N (%)
<b>Positive</b>	93(96.9)	81(84.4)	12(12.5)
<b>Negative</b>	03(3.1)	-	03(3.1)
<b>Total</b>	96(100)	81(84.4)	15(15.6)



**Figure 1.4: Bar diagram showing correlation between KOH & culture result.**

Out of total 81 culture positive samples, dermatophytes 63 (77.8%) were predominant, followed by *Candida* spp. 3 (3.7%) and only 1 (1.2%) non-dermatophyte. Among dermatophytes, *T. mentagrophytes* 25 (30.9%) was the predominant species, most commonly isolated form *T. corporis* 15 (18.5%), followed by *T. cruris* 5 (6.2%), *T. faciei* 4 (4.9%) and *T. unguium* 1 (1.2%). Second commonest isolate was *T. tonsurans* 17 (21%), with maximum isolation from *T. corporis* 11 (13.5%) followed by 2 (2.5%) cases each in *T. capitis*, *T. cruris* and *T. unguium*.

Out of total 7 (8.6%) *T. rubrum* isolates, 3 (3.7%) each were isolated from *T. pedis* and *T. manuum* and only 1 (1.2%) from *T. corporis* case. Five (6.2%) isolates of *M. audouinii* were from *T. capitis* 4 (4.9%) and *T. corporis* 1 (1.2%) cases.

All 3 (3.7%) isolates of *M. ferrugineum* and *T. schoenleinii* were isolated from *T. cruris* and *T. corporis* cases respectively. Two (2.5%) isolates of *T. verrucosum* were equally distributed in *T. capitis* and *T. unguium* cases. Lastly, only 1 (1.2%) isolate of *T. interdigitale* was isolated from a *T. manuum* case. 14 (17.3%) samples developed contamination while *Candida* spp. was isolated from 3 (3.7%) *T. corporis*, 2 (2.5%) and 1 (1.2%) *T. capitis* cases, while only one non-dermatophyte – *Fusarium oxysporum* was isolated from only 1 (1.2%) case of *T. unguium*. (Table 1.5)

**Table 1.5 Distribution of fungal isolates among various clinical types**

Clinic	Cultu	Dermatophytes	Non Dermatophytes
--------	-------	---------------	-------------------

al types	re Positive N (%)	<i>T. mentagrophytes</i>	<i>T. tonsurans</i>	<i>T. rubrum</i>	<i>M. audouinii</i>	<i>M. ferrugineum</i>	<i>T. schoenleinii</i>	<i>T. verrucosum</i>	<i>T. interdigitale</i>	Contaminant	<i>Candida spp.</i>	<i>F. oxysporum</i>
<b>T. corporis</b>	45(55.6)	15	11	1	1	0	3	0	0	12	2	0
<b>T. cruris</b>	10(12.4)	5	2	0	0	3	0	0	0	0	0	0
<b>T. capitis</b>	9(11.1)	0	2	0	4	0	0	1	0	1	1	0
<b>T. pedis</b>	4(4.9)	0	0	3	0	0	0	0	0	1	0	0
<b>T. manuum</b>	4(4.9)	0	0	3	0	0	0	0	1	0	0	0
<b>T. unguium</b>	5(6.2)	1	2	0	0	0	0	1	0	0	0	1
<b>T. faciei</b>	4(4.9)	4	0	0	0	0	0	0	0	0	0	0
<b>Total N (%)</b>	<b>81(100)</b>	<b>25(30.9)</b>	<b>17(21)</b>	<b>7(8.6)</b>	<b>5(6.2)</b>	<b>3(3.7)</b>	<b>3(3.7)</b>	<b>2(2.5)</b>	<b>1(1.2)</b>	<b>14(17.3)</b>	<b>3(3.7)</b>	<b>1(1.2)</b>

### V. Discussion

Superficial mycoses are one of the commonest skin diseases worldwide and are emerging as a major public health concern. Children are mostly affected because of predisposing factors such as poverty, overcrowding, and lack of guidance (Ogbu et al., 2015)<sup>4</sup>.

Out of total 96 cases of superficial mycoses, the most common clinical type was T. corporis findings are similar to previous studies. (Table 1.1)

**Table 1.1 Comparative analysis of commonest clinical types**

Commonest Clinical type	Present study	Dash et al., 2017 (Odisha) <sup>5</sup>	Fernandes et al., 2001 (Brazil) <sup>6</sup>
<b>T. corporis</b>	56.20%	47.40%	44.80%
<b>T. cruris</b>	13.50%	50%	56.90%

Out of the 81(84.4%) culture positive samples 65.6% males and 34.4% females were affected, with male female ratio of 2:1. The prevalence of our study is in accordance to previous studies conducted in Nigeria by Ogbuet al., 2015<sup>4</sup> and Adefemi et al., 2011<sup>1</sup>, who reported the similar prevalence in male and females of 61.3% & 38.7% and 68.4% & 31.6% respectively. However, Ogbu et al., 2015<sup>4</sup> & Oke et al., 2014<sup>7</sup> have reported equal prevalence in males and females of 56.5% & 43.5% Chukwu et al., 2011<sup>8</sup> from Nigeria. The lower incidence of females may be also due to the non-reporting to the hospital due to prevailing social stigma in the general population in males due to the higher exposures in the school and sporting activities and due to the wearing of uniform and closed footwear for prolonged periods in all weathers. This promotes sweating and sweats retentions due to high environment moisture content in summer season, thus facilitating fungal growth resulting in a high incidence of fungal diseases in males.

Dermatophytosis was most prevalent in the 11-14 year age group 67.7%, followed by 1-5 (15.6%), 11(11.4%), and 5(5.2%) in year age groups. Our findings are in accordance with previous studies by Dash et al., 2017<sup>5</sup> from Odisha. Only Oke et al., 2014<sup>7</sup> have reported highest prevalence of 49.7% in 7-11 year age group and Chukwu et al., 2011<sup>8</sup> have reported a prevalence of 41.0%. However, prevalence in age group 5-9 have been 40.7% reported by Ogbuet al., 2015<sup>4</sup> from Nigeria. Our findings confirm that dermatophytosis is a common finding in adolescent age group with minor variance from place to place.

On direct microscopic examination by KOH mount, out of the total 96 samples 93 (96.9%) samples were positive while only 3(3.1%) were negative. while 81 (84.4%) were culture positive and 12(12.5%) were negative. Out of 93 KOH positive samples 81(84.4%) samples culture positive while all 3 KOH negative, were reporting a high KOH and culture positivity, which moral boosting for an upcoming department like us. High KOH and culture positivity comparative has been reported by. (Table 1.2 & 1.3)

**Table 1.2: Comparative analysis of KOH**

KOH Results	Present study	Naglot et al., 2015 (Assam) <sup>9</sup>	Gupta et al., 2014 (Jaipur) <sup>10</sup>
Positive	96.90%	69%	74%
Negative	3.10%	30.80%	26%

**Table 1.3: Comparative analysis of culture result from previous studies**

Culture Results	Present study	Ayorinde et al., 2013 (Nigeria) <sup>11</sup>	Oke et al., 2014 (Nigeria) <sup>7</sup>	Chukwu et al., 2011 (Nigeria) <sup>8</sup>
Positive	84.4%	57.7%	64.1%	98.2%
Negative	12.5%	42.3%	35.9%	1.8%

In present study, *T. mentagrophytes* 30.9% was the predominant isolate followed by *T. tonsurans* 21%, *T. rubrum* 8.6%, *M. audouinii* 6.2%, *M. ferrugineum* 3.7%, and *T. interdigitale* 1.2%. We did not isolate any Epidermophyton species in this study. Previous studies have reported *T. mentagrophytes* as the predominant dermatophytic isolate from superficial mycoses cases in our region and India and our study confirms their findings. (Table 1.4)

**Table 1.4: Comparative analysis of commonest clinical isolates**

Clinical isolate	Present study	Chukwu et al., 2011(Nigeria) <sup>8</sup>	Ndako et al., 2012 (Nigeria) <sup>12</sup>	Arenas et al., 2004 (Mexico) <sup>3</sup>
<i>T. mentagrophytes</i>	30.9%	24%	2.20%	5.40%
<i>T. tonsurans</i>	21%	5.50%	1.10%	8.10%

## VI. Conclusion

Fungal infections of the skin and nails have been found in the last few decades to affect 20-25% of the world's population, making them one of the most frequent forms of infection. They represent a major public health problem in school age children especially in low- and middle-income countries (LMICs) like India where possible predisposing factors in acquiring the infection such as, poor hygiene, overcrowding, and low socioeconomic factors remain present. Laboratory diagnosis for superficial mycoses is variable and KOH mount microscopy and fungal culture is the most sensitive method available for its accurate diagnosis.

However, the present study is a small study that focuses primarily on the prevalence of different dermatophytes species in Northern Lucknow and a systematic study covering larger population and over a longer period of time would give a better insight into the epidemiology of dermatophytes in Lucknow and neighbouring region.

## Acknowledgments

The author would like to acknowledge the college of Integral Institute of Medical Sciences & Research, Integral University, and the end consumable materials for free. The author would like to express our sincere thanks and gratitude to respected guide Dr.SanjeevSahai (HOD), Department of Microbiology, IIMS&R and all faculties of Integral Institute, Lucknow.

### References

- [1]. Adefemi SA, Odeigah LO, Alabi KM. Prevalence of dermatophytosis among primary school children. Niger J Clin Pract 2011;14:1.
- [2]. Aggarwal A, Arora U, Khanna S. Clinical and mycological study of superficial mycoses in Amritsar. Indian J Dermatol 2002;47:218-220.
- [3]. Arenas R, Esmenjaud J R. Onychomycosis in childhood: A current perspective with emphasis on the review of treatment. An bras Dermatol, Rio de Janeiro 2004;79(2):225-232.
- [4]. Ogbu CC, Okwelogu IS, Umeh AC. Prevalence of superficial fungal infections among primary school pupils in Awka South Local government area of Anambra State. JMR 2015;2(1):15-22.
- [5]. Dash M, Panda M, Patro N, Mohapatra M. Socio demographic profile and pattern of superficial dermatophytic infections among pediatric population in a tertiary care teaching hospital in Odisha. Ind. J.Paediatr.Dermatol 2017;18:191-5.
- [6]. Fernandes NC, Akiti T, Barreiros C. Dermatophytoses in children: Study of 137 cases. Rev. Inst. Med. Trop. S. Paulo 2001; 43(2): 83-85.
- [7]. Oke OO, Onayemi O, Olasode OA, Omisore AG, Oninla OA. The Prevalence and Pattern of Superficial Fungal Infections among School Children. Dermatolo Res Pract 2014;842917: 7.
- [8]. Chukwu ID, Chukwu OOC, Chuku, Enweani IB. Dermatophytoses in rural school children associated with livestock keeping in Plateau State, Nigeria. J. Yeast Fungal Res 2011;2(1): 13-18.
- [9]. Naglot A, Shrimali DD, Nath BK, Gogoi HK, Veer V, Chander J, Tewari R. Recent trends of dermatophytosis in Northeast India( Assam) and interpretation with published studies. Int. J.Curr.Microbiol. App.Sci 2015;4(11): 111- 120.
- [10]. Gupta S, Agrawal P, Rajawat R, Gupta S. prevalence of dermatophytic infection and determining sensitivity of diagnostic procedures. Int. J. Pharm.Sci 2014;6(3): 35-38.
- [11]. Ayorinde AF, Adesanya OO, Alaran OA. A Microbiological Study of Dermatophyte Infection among Primary School Children in Mowe. Curr. Res. J. Biol. Sci 2013;5(5): 205-209.
- [12]. Ndako JA, Omorefosa O, Olopade B. prevalence of dermatophytes and other associated among school children. Res. J. Med. Med. Sci 2012;1(3): 049-056.

Alok Kumar, et al. "A Study of Fungal Isolates from Superficial Mycoses Cases in Pediatric Patients, Lucknow." *IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)*, 19(4), 2020, pp. 07-13.