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Epidemiologic study of Dermatophytosis in Al-Najaf government

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Abstract
Dermatophytosis, commonly referred to as ring worm which, is a superficial fungal infection caused by filamentous fungal species (Dermatophytes) belonging to three anamorphic genera, ***Trichophyton sp.***, ***Microsporum sp.*** and ***Epidermophyton sp***. It is the most common superficial fungal infection in the world. Due to the importance of high prevalence rate of above diseases, this study was conducted to determine the epidemiologic condition of diseases in Al-Njaf province. Total of 216 specimens of (skin scrappings, hair fragments and nail clippings) were collected. Also 138/216 (63.9%) specimens exhibited positive results in both direct examination and culture, and 32/216 (14.8%) specimens were negative in direct examination and positive in culture therefore only 170/216 (78.7 %) specimens yielded growth of dermatophytes isolates and considered as positive specimens. ***Microsporum canis*** showed highest frequency in 54/216 (31.8 %) isolates, while ***M. audouinii*** and ***Trichphyton schoenleinii*** in 8/216 (4.71%) isolates showed lowest frequency. Also 38/216 (22.4%) of ***M. canis*** isolates were in male while 16/216 (9.41%) isolates in female, both ***T. schoenleinii*** and ***M. audouinii*** were not recorded in female 0(0%). Tinea corporis was the predominant infection in 75/157 (47.77%) patients followed by Tinea capitis in 29/157(18.47%) patients. Gender is not statically significant with dermatophytosis, while a significant ($P<0.01$) correlation was observed with age, residency variables. Age revealed significant effect ($P<0.05$) on the distribution of dermatophytosis patients**,** the age bracket 1-10 years were highest frequency with 42/157(26.8%) patients, while age bracket 61-70 years were lowest frequency 2/157(1.3%) patients. Residency revealed non significant association in relevance with type of dermatophytosis, while significant ($P<0.01$) correlation was observed with gender. This study showed that there is a high prevalence rate of Dermatophytosis in Al-Najaf province. Therefore, due to importance of the disease, it is necessary to diagnose and cure the disease immediately.

Introduction
Dermatophytosis is referred to all fungal infections of skin that caused by different species of dermatophytes [1]. Other accountable species for dermatophyosis are yeast and yeast like fungi (ie, ***Candida***, ***Geotrichum candidum***, ***Trichosporon beigelli***) and saprophytic fungi such as ***Aspergilus***, ***Scopulariopsis***, ***Fusarium***, ***Acremonium*** and ***Penicillium*** [2]. It is estimated that dermatophytosis are including 73.5% of 4000 fungal infections [3] . Fungal infections are not confined to developing countries and they are seen even in developed countries as USA. The prevalence rate of the disease is so high in USA has that the expense of not fungal agents used for skin infections amounted to 409 million dollars during 16 years (25 million dollars yearly) **[4]**. Due to its hot and humid climate and specific environment, existing some jobs such as animal husbandry with non-hygienic methods, which make direct contact of human to animals, and the unfamiliarity with how the disease may be acquired. Iraq is among the countries where dermatophytosis is mostly prevalent **[5]**. The aim of present

study was to determine the epidemiologic condition of dermatophytosis in Al-Najaf province .

Materials and methods

A total of 216 specimens were collected from 157 patients with dermatophytosis which were clinically diagnosed by Dermatologist, were recruited in the current study after attending to the consultant of Dermatology and Venereal Disease in Al-Sadder Medical City in Al-Najaf Government , from January 2011 to November 2011.

Collection of specimens:

Skin scrapping were collected by disposable scalpel blades of the solid type held vertically to the skin . Cleaning of the skin with alcohol was done if the patient has applied ointment or powder. If the lesion has a definite edge, the material was taken from the active margin also transport swabs were used as a good back-up tools. When blisters are present, a pair of fine scissors was used to cut off a blister roof for microscopic examination and culture. The scrapings were collected and transported in folded paper, which keeps the specimen dry, thus preventing over-growth of bacterial contaminants, squares of dark colored paper card are ideal; and then be carefully folded and secured by a paper clip. Hairs to be examined for dermatophytosis if suspected. The hairs were removed with the roots intact; cut hairs are unsuitable. In cases of kerion, a transport swab wiped over the lesion were usually picked up enough conidia to give a positive culture. The full thickness of the nail were sampled, debris from under the nail was collected because it is a fruitful source of material, and scraped out using the flat end of a dental probe.

Direct examination:

For routine examination, specimens were mounted in 10 % KoH; For skin scrapings and nails, warming gently over the burner was done to speed up the process, but boiling was avoided. A few minutes spent carefully preparing the specimen in this way. Nail specimens was take longer to clear than skin (in some case specimen incubated in 37º C for hours), but small pieces and debris are taken, they soften within 10 min. In contrast to skin and nail specimens, infected hairs are very delicate, and if heated or left in mounting fluid for more than a few minutes tend to disintegrate, obscuring the characteristic arrangement of the arthroconidia. Methylene blue or lactophenol cotton blue stains must be used to enhance the contrast between fungus and skin.

Culture of Specimens

The primary culture media were (SDA) and (PDA) with addition of 1 g/l cyclohexamide to inhibit the growth of non-dermatophytes moulds and 0.05 g/l chloromphenicole to reduce contamination with fast growth Bacteria. Specimens were cultured on the disposable Petri dishes (duplicate) filled with 15 ml of medium were incubated at 29±2ºC for 14-21 days. Petri dishes were examined daily after 7 to14 days [6].

Identification of Dermatophytes isolates

The following criteria were taken in consideration in the growth identification: first, Morphological features of growth colony which were including (color, texture, margin, consistency, colony reverse and pigments that reproduced). Second, Microscopical and Macroscopical features (size, shape, arrangement, of

microconidia and macroconidia, their conidial ontogeny, types of specialized structure upon which spores were borne) [6,7,8,9,10,11].

Results

1-Patients

The present study included a collection of 216 specimens from the randomly recruited 157 patients which include 123(56.94%) skin scrapping specimens 81(65.85%) were collected from male and 42(34.15%) were obtained from female, while total of 61(28.24%) hair fragments specimens 13(21.31%) were collected from male and 48(78.7%) were collected from female and finally 32 (14.81%) nail clipping specimens distributed 8(25%) , 24(75%) specimens for male and female respectively (Table 1).

Table (1): Numbers and percentages of specimens and gender of patients with dermatophytosis in Al-najaf government.

Type of specimens	No. of specimens (%)	No. of Male (%)	No. of Female (%)
Skin scrapping	123(56.94%)	81(65.85%)	42(34.15%)
Hair fragments	61(28.24%)	13(21.31%)	48(78.7%)
Nail clipping	32(14.82%)	8(25%)	24(75%)
Total	216(100%)	102(47.22%)	114(52.78%)

2-Isolation:

The results also revealed that 138/216 (63.9%) specimens were positive in both direct examination and culture, 21/216 (9.7%) specimens has been shown positive in direct examination and negative in culture, and 32/216 (14.8%) specimens were negative in direct examination and positive in culture, while the cases of negative results in both direct examination and culture were 25/216 (11.6 %) specimens (Table 2).

Table (2): Numbers and Percentages of diagnostic specimens by direct examination and culture method collected from patients with dermatophytosis in Al-najaf government.

Clinical cases examination	Number	Percentage
Positive in both direct examination and culture	138	63.9%
Positive in direct examination and Negative in culture	21	9.7%
Negative in direct examination and Positive in culture	32	14.8%
Negative in both direct examination and culture	25	11.6 %
Total number	216	100%

The current study revealed that out of 61 cases that were a positive on direct microscopic examination, the invasion of hair was of ectothrix type, forming masses of arthroconidia on the outside of the hair shaft in 39 (63.9%) specimens,

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while the invasion of hair was endothrix type, and abundant sporulation inside the hair shaft causes breakage of the hair near the surface of the scalp in 22 (36.1%) specimens (Figure 1).

Out of a total of 216 investigated specimens only 170/216 (78.7 %) yielded growth of dermatophytes isolates were cultured on SDA, and considered as positive specimens which were used in the phenotypic and genotypic diagnosis (Table 3). Nine of dermatophytes species were identified in specimens of examined patients, out of them *M. canis* in 54(31.8 %) isolates showed the highest frequency followed by *T. mentagrophytes* 38(22.4%) isolates, *T. tonsurans* 18(10.6%) isolates, *E. floccosum* 14(8.24%) isolates, *T. rubrum* , *T. ajelloi* and *M.gypseum* 10(5.88%) isolates and finally *M. audouinii* and *T. schoenleinii* 8(4.71%) isolates. In brief, the following observations were made: of the 216 specimens, 110/170(64.7%) isolates were collected from male, and 60/170 (35.3%) isolates were recovered from female, also the present study revealed that *M. canis* isolates was much more common in male 38(22.4%) than in female 16(9.41%) while the both *T. schoenleinii* and *M. audouinii* were not recorded in female 0(0%) .

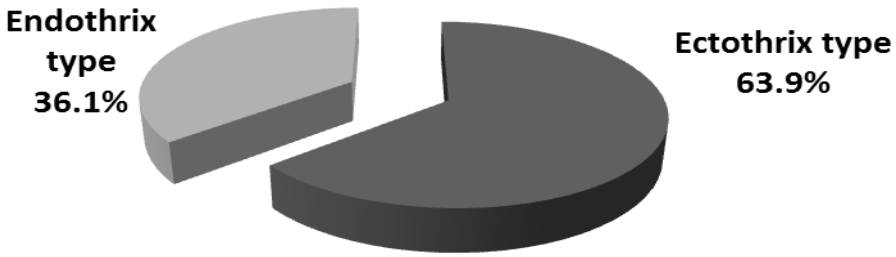

Figure (1): Types of hair infection of dermatophytes .

Table (3): Dermatophytes species isolated from 157 patients with dermatophytosis in Al-najaf government .

Dermatophytes species	No.(%)	Male(%)	Female(%)
M. canis	54(31.8)	38(22.4)	16(9.41)
T. mentagrophytes	38(22.4)	26(15.3)	12(7.06)
T. tonsurans	18(10.6)	12(7.06)	6(3.53)
E. floccosum	14(8.24)	4(2.35)	10(5.88)
T.ajelloi	10(5.88)	6(3.53)	4(2.35)
T. rubrum	10(5.88)	4(2.35)	6(3.53)
M.gypseum	10(5.88)	4(2.35)	6(3.53)
T. schoenleinii	8(4.71)	8(34.71)	0(0.00)
M. audouinii	8(4.71)	8(4.71)	0(0.00)
Total	170(100)	110(64.7)	60(35.3)

3- Clinical Types of dermatophytosis:

Tinea corporis was the predominant infection which recorded in 75 (47.77%) patients followed by, Tinea capitis 29(18.47%), Tinea pedis 12 (7.64%), Tinea unguium16 (10.19%), Tinea manuum 8 (5.10%), Tinea cruris 7 (4.46%) and Tinea faciei 10(6.37%) of patients respectively (Figure 2).

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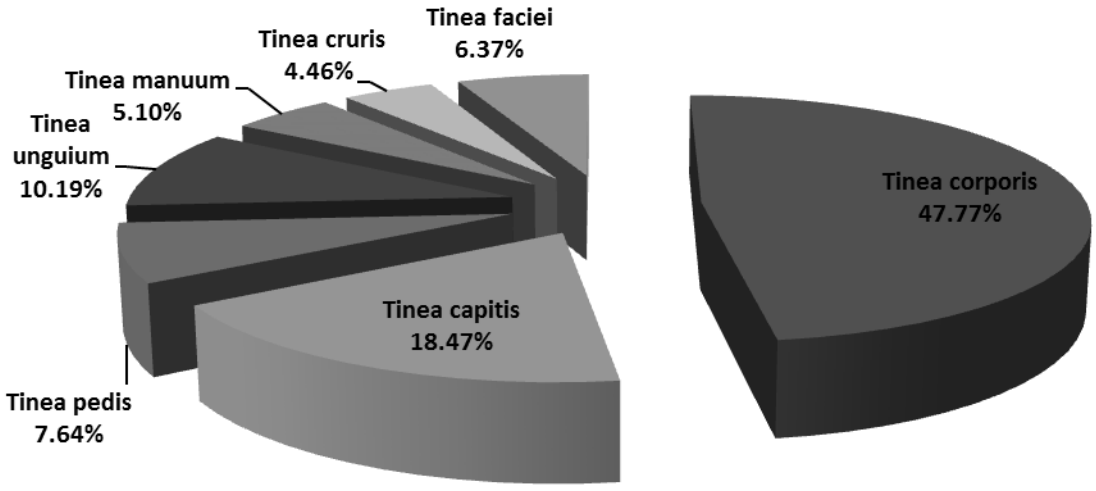

Figure (4): Percentages of infection in clinical types of dermatophytes

Relationship between dermatophytic infection and anthropometric parameters, and other variables .

The gender relationship of 157 patients were 89/157 (56.69%) males and 68/157 (43.31%) females. Gender relationship is not statically significant, while a significant ($P<0.01$) correlation was observed with age, residency, occupation and presence of chronic disease variables. Patients with Tinea corporis recorded the highest frequency 75/157 (47.77%) patients divided in to 42/157 (26.75%) male and 33/157 (21.02%) female (Table 4 and 7). According to patients age it was found that statistically significant effect ($P<0.05$) on the distribution of dermatophytosis patients, results exhibited that the age bracket 1-10 years had the highest frequency with a total 42/157(26.8%) patients, ages 21-30 years were observed to be at the second rank in the province 36/157 (22.9%) patients, while age group (months) and (61-70) years recorded the lowest frequency 4/157(2.6) patients and 2/157(1.3%) patients respectively. However other age related groups illustrated varied distributions. Significant correlation ($P<0.05$) was observed with type of clinical types of dermatophytosis especially with Tinea capitis and Tinea corporis 20/157(12.7%) cases and 19/157(12.1%) cases in age bracket 1-10 years old respectively, while a significant correlation at ($P<0.01$) level was observed with (gender, occupation, domestic animals existence and presence of chronic disease (Table 5 and 7).

The influence of residence on the distribution of the patients was evaluated. 120/157 (76.43%) patients was from urban area while 37/157 (23.57%) patients from rural areas, both group were demonstrate to be non significant effect in relevance with type of dermatophytosis. The study showed that the tinea corporis had the highest frequency of dermatophytosis types with a total of 75/157(47.77%) cases, urban area 50/157(31.85%) cases and rural area was 25/157(15.92%) cases. In other hand a significant ($P<0.01$) correlation was observed with (gender, existence of domestic animals, presence of chronic disease and usage of treatment) variables (Table 6 and 7).

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Table (4): Distribution of patients with dermatophytosis according to gender.

Clinical types		Gender		
		Male	Female	Total
Tinea corporis	No.	42	33	75
	%	26.75	21.02	47.77
Tinea capitis	No	23	6	29
	%	14.65	3.82	18.47
Tinea pedis	No	1	11	12
	%	0.64	7.01	7.64
Tinea unguium	No	3	13	16
	%	1.91	8.28	10.19
Tinea manuum	No	6	2	8
	%	3.82	1.27	5.10
Tinea cruris	No	7	0	7
	%	4.56	0	4.46
Tinea faciei	No	7	3	10
	%	4.46	1.91	6.37
Total	No.	89	68	157
	%	54.7	43.31	100

Table(5): Distribution of patients with dermatophytosis according to age

Clinical type		Age (year)								
		Month	1-10	11-20	21-30	31-40	41-50	51-60	61-70	Total
Tinea corporis	No.	4	19	17	15	13	7	0	0	75
	%	2.6	12.1	10.8	9.6	8.3	4.5	.0	.0	47.77
Tinea capitis	No.	0	20	4	3	2	0	0	0	29
	%	.0	12.7	2.6	1.9	1.3	.0	.0	.0	18.47
Tinea pedis	No.	0	0	0	3	1	2	4	2	12
	%	.0	.0	.0	1.9	0.6	1.3	2.6	1.3	7.64
Tinea unguium	No.	0	0	1	5	3	4	3	0	16
	%	.0	.0	0.6	3.2	1.9	2.6	1.9	.0	10.19
Tinea manuum	No.	0	0	1	6	1	0	0	0	8

	%	.0	.0	0.6	3.8	0.6	.0	.0	.0	5.10
Tinea cruris	No.	0	0	0	1	4	2	0	0	7
	%	.0	.0	.0	0.6	2.6	1.3	.0	.0	4.46
Tinea faciei	No.	0	3	3	3	0	1	0	0	10
	%	.0	1.9	1.9	1.9	.0	0.6	.0	.0	6.37
Total	No.	4	42	26	36	24	16	7	2	157
	%	2.6	26.8	16.6	22.9	15.3	10.2	4.5	1.3	100

Table(6): Relationship between dermatophytes infection with localities of the patients .

Clinical type		Residency		
		Urban	Rural	Total
Tinea corporis	No.	50	25	75
	% o	31.85	15.92	47.77
Tinea capitis	No.	22	7	29
	%	14.01	4.46	18.47
Tinea pedis	No.	12	0	12
	%	7.64	0	7.64
Tinea unguium	No.	16	0	16
	%	10.19	0	10.19
Tinea manuum	No.	5	3	8
	%	3.19	1.91	5.1
Tinea cruris	No.	5	2	7
	%	3.19	1.27	4.46
Tinea faciei	No.	10	0	10
	%	6.37	0	6.37
Total	No.	120	37	157
	%	76.43	23.57	100

Table (7):Statistical correlation between the various variables in the present study

Variables		Gender	Age	Residency
Gender	r-value	1	0.26**	0.21**
	p-value		0.001	0.004
Age	r-value	0.264**	1	0.119
	p-value	0.001		0.073
Residency	r-value	0.216**	0.119	1
	p-value	0.004	0.073	
	p-value	0.001	0.001	0.005

*(P<0.05),** (P<0.01)

Discussion:

Infections caused by dermatophytes are widespread, are increasing in prevalence on a global scale, and have been considered a major public health concern in some areas of the world, which accounts for as many as 69.5% in humans [12]. In addition to the well known superficial infections caused by this organism, such as Tinea capitis, Tinea corporis, Tinea nails, Tinea unguium, and Tinea pedis, dermatophytes species is also responsible for deep dermal invasion in immunocompromised patients . Moreover, dermatophytosis infections are often intractable, and relapses frequently occur after cessation of antifungal therapy [13].

Furthermore dermatophytes are parasitic fungi that infect skin, hair and nails of both humans and animals. They are the primary causative agents of dermatophytosis, a major public health concern in some geographic regions [14,15].

While not fatal, dermatophyte infections cause significant morbidity and are of significant cost to society because of their chronic nature and resistance to therapy. Dermatophytes encompass three genera, the following three species , ***T. rubrum***, ***T. mentagrophytes*** and ***M. canis*** are the most common species in hospital isolates (72–95%) [16,17]. Dermatophytosis has been reported to be encouraged by hot and humid conditions and poor hygiene and occur throughout tropical and temperate regions of the world [18].

One aim of this investigation was to evaluate the spread of dermatophytes isolates and to ascertain the detection rate of these microorganisms in dermatophytosis patients in Al-Najaf government, since the present study focused on detection of various clinical cases of dermatophytosis and deferent dermatophytes isolates.

During the period of present study, the results of direct examination revealed that it was very essential, as it allows the clinician to start treatment, pending culture. Although in the present study false-negative results in both direct examination and culture have been reported in 25 (11.6 %) specimens in routine practice, these are depending essentially on the skill of the observer and on the quality of sampling. Cultures remain negative in spite of the positivity of direct examination in 21(9.7%), therefore out of the positive specimens by culture 138 (63.9%) were positive by direct KOH microscopic examination. This was in relatively agreement with Escobar and Carmona-Fonseca(2003)[18], Arca *et al* (2004)[19] and Samia *et al* (2006)[20] who found that direct microscopy was positive in 92%, 77% and 71.7% respectively from all culture positive cases.

In this study the direct microscopic examination shows false negative results in 57(26.4%) of specimens, this is relatively in accordance with Liu *et al* (2000)[21] who stated that this method is insensitive, showing false negative results up to (19%) while discordance with Tampieri (2004)[22] who mentioned that direct microscopic examination shows false negative results up to (50%). These false-negative results may be related to an insufficient amount of material or a specimen poor in fungal elements, but also to a too short incubation time, a non-suitable temperature or the presence of ‘‘contaminants’’ which can prevent the development of the pathogen. False-negative results on Sabouraud’s agar may also result from an antifungal treatment initiated before sampling. Many antifungal drugs currently used for treatment of dermatophytosis are retained for a long time within the horny layer of the epidermis, and drug residues in the sample may inhibit the growth of the pathogen [23,24,11,9,25].

The emergence of negative results by culture, may be also caused by an error in the method of sample storage until culture , as stored in containers which will help preserve moisture in the growth of saprophytic fungi which contaminate the pitch original sample when culture and then do not see a positive result is culture (Al-Hashemy, 1979)[26].

The outcome invasion of hair by ectothrix type were (39) (63.9%) specimens, while those of endothrix type invasion were (22) (36.1%) specimens . It has been shown from the foregoing results that the endothrix type invasion was the prevalence type of hair infection, and this result is consistent with [27,28,29], and discordance with the results of [30,31,32].

Dermatophytes identification

Conventional laboratory identification of dermatophytes still consists primarily of micro- and macro-morphological examination of primary isolates, supplemented with physiological tests for atypical isolates in some laboratories in wealthier parts of the globe, the diagnostic therefore, tends to be stripped down to the minimal cost necessary to achieve targeted proficiency levels that are based mainly on the ability to identify typical isolates accurately [33].

The strong point of this approach is that the great majority of dermatophytes are typical in outgrowth, and can be readily identified based on conidia, hyphal features, and colony appearance as seen in primary culture on SDA and PDA [34,35]. Therefore as we found in present study there was only 170 (78.7 %) out of 216 specimens of patients revealed successful cultivation, and considered as positive specimens which were used in the phenotypic and genotypic diagnosis. Nine dermatophytes species were identified out of which ***M. canis*** 54 (31.8%) isolates showed the highest frequency from total summation of dermatophytes isolates followed by *T. mentagrophytes* 38 (22.4%) isolates, and both ***T. schoenleinii*** , ***M. audouinii*** 8 (4.71%) isolates were appeared in lowest frequency of dermatophytes, these results were agreement with Al-Temimi,2003 in Al-najaf government.

On other hand the agreement documented results in pervious researches [36,37,38], revealed that *M. canis* is the main causative agents of dermatophytosis in these countries. Also present results study were discordance with the results of Ali (2009) in Al-Qadsiya government[39] , Abbas (1995)[28], Ali(1990)[27], Al-Janabi (2006)[40] , Hassan (2007) in Baghdad[29], and Al-Hazaaly (2005) in Baquba[41], which were showed that ***T.mentagrophytes*** revealed highest frequency from total summation of dermatophytes isolates. [42], was found that ***T. verrucosm*** isolates was the main causes of dermatophytosis . [43,30,31,44] were observed that ***T. verrucosm*** is responsible for most cases of dermatophytosis.

The variation of occurrence results of isolates may be due to differences in location of patients residency furthermore type of clinical case. The types of skin fungi vary from one place to another, also it is depending on the ability of these fungi dissemination, some of them have dissemination all over the world and each other, spreading in certain areas and this leads to infection of differences from place to place [45,33].

Clinical Types of dermatophytosis

The present study pointed out prevalence of Tinea corporis in the first rank 70 (46.7%) patients, and Tinea capitis in the second rank 27(18%) patients in investigated patients followed by other clinical types of dermatophytosis . The comparison of the present distribution of patients with others mentioned in the literature highlights different observations, [20] have found that a distribution of 47(41%) cases of Tinea capitis and 29(25.30) cases of Tinea corporis, [43] who emphasized that Tinea capitis is considered the major type of fungal infection in Egypt, in Libya Tinea corporis accounted for 45.9% cases followed by Tinea capitis as discussed by [46], while in Yemen Tinea corporis accounts for the majority of cases followed by Tinea capitis as mentioned by[47].

It seems that the distribution of patients was varied, and depended on the selection of patients and/ or the host and environmental factors, furthermore the importance of regional variation .

Relationship between dermatophytic infection and anthropometric parameters, and other variables .

In present study we found that non significant effect in both 89/157 (56.69%) males and 68/157 (43.31%)females on the distribution of patients with different clinical types of dermatophytosis, while a significant ($P<0.01$) correlation was observed with (age, residency, occupation and presence of chronic disease variables). Gender related distribution of dermatopytosis diagnosed patients was investigated in previous reports, some of them have mentioned gender differences, while others did not mention such differences.

Our present results were disagree with several findings such as, [27,28, 42, 40, 29, 48, 49, 50, 51, 52] shows that boys are most susceptible than girl to dermatophytes infection especially infection of the scalp. The higher susceptibility of boys could be due to their frequent contact with soil as a result of farm work and football.

Several studies have highlighted a decreased prevalence in male patients relative to those of females also these results were not coincided with our results, [53] in Brazil described that Tinea affects females more than males and this may be due to differences in the results of healthy habits and personal hygiene which is one of a many factors influencing the occurrence of Tinea. Some reports have failed to demonstrate gender difference in the distribution of patients. [41] have suggested no difference between male and female prevalence of patients with various clinical types of dermatophytosis, therefore compatible observations have been indicated by our present study .

Age was found to have an effect on the distribution of patients with different clinical types of dermatophytosis; patients of ages 1-10 years old exhibited highest percentage 42/157(26.8%) patients among age related groups. Patients of ages 21-30 years old were observed to be at the second rank 36/157 (22.9%), while age groups (months) and (61-70) years old recorded the lowest frequency 4/157(2.6)

and 2/157(1.3%) respectively, a significant difference (P<0.05) was observed with type of clinical types of dermatophytosis.

Inconsistency was observed when the current results were compared with some mentioned previously in the literatures, the results in the present study was in agreement with [48] who found prevalent infection especially among children below 10 years old of age, and the results of [51], that Tinea infection was more frequent among children between ages 5 and 10 years old. while the discordance results by [54, 55,56], who mention that the ages of the infected patients were between (2-14) years, furthermore a significant correlation at **(**P<0.01**)** level was observed with (gender, occupation, domestic animals existence and present of chronic disease). I think that the differences in the incidence of ringworm infection between the age groups and sexes seem, in general, to reflect differing rates of exposure and of sebum production, differing clothing and fluctuations of immunity with old age. Also the activity of saturated fatty acids with chain lengths of 7, 9, 11 and 13 carbon residues. It has been postulated that their presence on the skin in post pubertal children may account for the spontaneous resolution of Tinea capitis after this age, and the rarity of new infections in adults.

Otherwise the influence of residence on the distribution of the patients was evaluated. Patients of urban 120/157 (76.43%) cases and rural 37/**157** (23.57%) cases area were demonstrate to be non significant effect in relevance with type of dermatophytosis. This results were accordance with [57,42, 58] who mention that the urban area patients were much more than rural area patients, while the discordance results by [59,29, 39] who mention that the rural area patients were much more than urban area patients.

Our explanation for this results are overcrowding of population in urban leading to contact between healthy and patients either within their family or at school or at market, also the upbringing of domestic animals and other reasons such as living conditions, large family size and close contact, either directly or by sharing facilities, including combs and towels, is common between family members in low socioeconomic strata. All these reasons clarify the elevation of urban patients attribution. In other hand a significant (P<0.01) correlation was observed between residency and gender.

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