Histopathology of human scabies in Iraq

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Abstract:

This research was conducted in order to detect the histological alterations in the skin of scabies patients and to identify the etiological agent of mange using a genetic approach due to the significance of this zoonotic illness. This research was carried out in Iraq's Baghdad Province. In all, 327 scabies-infected individuals (both male and female) were included in this research between the beginning of January and the end of December 2022. Age ranges between 1 year and more than 50 years; gender; and presence of animals were among the data collected from patients. The diagnosis of scabies caused by Sarcoptesscabiei was established through physical examination, inspection of the affected skin area, or by extracting S. scabiei using a needle and obtaining a tissue sample for microscopic analysis to identify the presence of scabies mites or their eggs. The specimens were placed in sterile plastic containers and subsequently preserved in 100% ethanol. These samples were then stored individually at a temperature of 4 °C until DNA extraction. The manufacturer's instructions were followed while utilising a kit for DNA extraction. Two primers were used in this study for confirm diagnosis of infection which were COX and ScaBi. With a 4mm punch and 2% xylocaine as the anesthetic, skin biopsies were collected. The biopsies were regularly processed, embedded in paraffin slices, stained with hematoxylin and eosin, and viewed under a microscope after being fixed in 10% formalin. The current results showed that age group at 21-50 years were significantly infected than another age groups. The present results exhibited that males were significantly highly infected than females. The results displayed that infections in winter season were increased significantly than other seasons of the year. The molecular results showed that all samples were Sarcoptesscabie by using the two primers (COX) and ScaBI. Histopathological finding showed that the epidermis is thick, and the dermis has a minor buildup and infiltration of inflammatory cellsstratum corneum thickening and parasite presence were present.

Inconclusion, the findings of the present investigation indicate that scabies remains a neglected public health concern, with a significant number of individuals afflicted by this condition, resulting in various histological alterations and harm to the integumentary system.

Keywords: Histopathology, Molecular, age, gender, Sarcoptesscabie

Introduction:

The ectoparasite*Sarcoptesscabiei* is the source of the infectious skin condition known as scabies. One of the most overlooked health issues in the world is this illness. Few

research has been done regarding it, particularly in the third world countries, despite the fact that it infects more than 300 million people annually around the globe [1,2,3].

Mites itch so badly that people scratch their skin, which allows bacteria to get into the crusty portions of the skin and causes secondary infection [4]. Members of the same family who share linens and towels are contagious. Skin-to-skin contact and sexual activity are other ways that the illness may spread [5].

Both sexes and people of all ages may have scabies. It does not make a distinction between people's socioeconomic statuses and impacts all ethnic groups equally. Scabies is also a major issue for farmers and others who produce cattle, leading to large financial losses [6, 7].

The clinical manifestations of the illness may vary depending on factors such as the parasite load, the age of the host, the patient's immune status, and their general health [8]. Important signs of the illness in humans and other animals include the development of channels as well as tunnels in the skin, localised inflammation of the affected regions, and excruciating itching [9, 10].

This research was conducted in order to detect the histological alterations in the skin of scabies patients and to identify the etiological agent of mange using a genetic approach due to the significance of this zoonotic illness.

Materials and Methods:

This research was carried out in Iraq's Baghdad Province. In all, 327 scabies-infected individuals (both male and female) were included in this research between the beginning of January and the end of December 2022.

Age ranges between 1 year and more than 50 years; gender; and presence of animals were among the data collected from patients.

The diagnosis of scabies caused by Sarcoptesscabiei in patients was conducted through physical examination, inspection of the affected skin area, or extraction of a S. scabiei using a needle and subsequent removal of a small skin piece for tissue sampling. The tissue sample was then examined under a microscope to identify the presence of scabies mites or their eggs [11].

Patients with scabies have been isolated with the Sarcoptes mites. According to [12], The specimens were individually placed in sterile plastic containers and subsequently preserved in 100% ethanol. The samples were then stored separately at a temperature of $4 \,^{\circ}$ C until DNA extraction was performed.

The manufacturer's instructions were followed while utilising a kit (geneaid, Taiwan) for DNA extraction.

The primers were made in accordance with the manufacturing instructions (Table 1).

Gene	F	R	Amplic	Re
name			on size	f.
			(bp)	
COX	5'-		250	13
	CTTATTATTCCTGGAT	5'CTAATTTTCCTCCTAATAT		
	TTGGRTA -3'	TGTWGA -3'		
ScaBI	5'-	5'-	289	14
	TCTTAGGGGCTGGAT	GAAGCTTTTCACCATTAGA		
	TTAGTATG -3'	AGCTG		

Table 1. Primers in this study

After positioning the comb properly and pouring the agarose gel solution onto the tray, the comb was gently removed from the tray and 6 l of PCR product and 2 μ l of Ladder were added to each comb well. The comb was then permitted to solidify for 15 minutes at room temperature. An electric current was run for 30 minutes at 100 volts, then for 45 minutes at 50 volts.

Skin biopsies were obtained using a 4mm punch and 2% xylocaine as the local anaesthetic. The specimens underwent routine processing, were embedded in paraffin blocks, subjected to hematoxylin and eosin staining, and subsequently examined under a microscope following fixation in a 10% formalin solution.

Statistical analysis was done by using SPSS version 23.

Results and Discussions:

The current results showed that age group at 21-50 years were significantly infected than another age groups (Table 2).

Age groups (years)	Number	Percentage	P value
1-10	24	7.3	
11-20	71	21.7	
21-50	167	51.1**	0.04
More than 50	65	19.9	
Total	327	100%	

Table 2. Distribution according to the age

The present results exhibited that males were significantly highly infected than females (Table 3).

Gender	Number	Percentage	P value
Male	192	58.7**	0.05
Female	135	41.3	
Total	327	100%	

Table 3. Distribution according to the gender

The results displayed that infections in winter season were increased significantly than other seasons of the year (Table 4).

Season	Number	Percentage	P value
Winter	98	30**	0.06
Spring	87	26.6*	0.04
Summer	59	18	
Autumn	83	25.4*	0.03
Total	327	100%	

Tuble 4. Distribution of infection according to the season	Table 4.	Distribution	of infection	according	to the seaso
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The molecular results showed that all samples were Sarcoptesscabie by using the two primers (COX) (fig.1) and ScaBI (Fig. 2).



Figure 1: A PCR result of primer (COX) gel electrophoresis showing 250bp visualised under ultraviolet light after ethidium bromide staining.



Figure (2): 289bp Primer TM at $(59^{\circ}C)$ is seen in the gel electrophoresis for the PCR result of the Scabie primer. (Agarose 1%, 10 min. at 100 volts, then decreased to 70

volts, 60 min.visualised under ultraviolet lighting following ethidium bromide staining. DNA ladder, Lane L (100–1500 bp).

The epidermis is thick, and the dermis has a minor buildup and infiltration of inflammatory cells (Figure 3). Figure 4 showed the thickened epidermis with inflammatory cells aggregations. The stratum corneum thickening and parasite presence were present (Figure 5).



Figure 3. Scabies patient's epidermis demonstrating a mild buildup and infiltration of inflammatory cells in the upper dermis. (H&E,10x).





Figure 4. shows the thickening of the epidermis and the infiltration of inflammatory cells in layer (c) of the dermis. (H&E,10x).

Figure 5. stratumcorneum thickening and parasite presence were present(H&E,10x).

Discussion:

The current results showed that age group at 21-50 years were significantly infected than another age groups.

This outcome was consistent with [15] findings in Iraq, which indicated that 42 patients (31.8%) who had scabies were in the 30- to 44-year-old age range. The age group of 11 to 20 years old had the greatest percentage of scabies patients in Thiqar province (22.9%), while the age range of 81 to 90 years old had the lowest percentage of cases (0.16%). These findings were previously documented in article by [16].

According to Al-Musawi et al. (17), the age group of 6-20 years experiences the highest impact, with the age group of 10-19 years being affected by 41.7%. This is consistent with the findings of AlDulaimi and Al-Shaha (2018) reported elevated infection rates among specific age groups in Al-Anbar province. These age groups included preschool-aged children, individuals between the ages of 18-24, and those aged 30-36. The observed infection rates were 18.40%, 15.50%, and 13.80%, respectively.

The present results exhibited that males were significantly highly infected than females.

These findings are consistent with some earlier research conducted in Iraq. Al-Samarai (15) found that of the 11194 scabies cases he examined, 13.5% were males

and 8.6% were women. Another research conducted in Baghdad by Sharquie[19] revealed that out of a total of 97 individuals infected with scabies, 58 (59.8%) were men and 39 (40.2%) were women. [13]discovered 1112 patients in the Al-Najaf province, of whom 602 (54.1% of men) and 501 (45.9% of women) were patients. Alzobydy[20] discovered in his research (344) patients in Baquba City who were diagnosed with scabies (2.16%); there were 130 (37.3%) girls and 214 (62.2%) men. By Mousa and Hassan [16], who established the province of Thiqar, the high percentage of scabies sufferers were men, 55.9%, and females, 44.1%.

Historically, scabies has been thought to spread more easily in the winter than the summer [21; 22; 16] due to the increased use of blankets and other coverings during cold weather. Nonetheless, the present investigation revealed that the incidence of infection was comparable during both the cold and warm seasons. The high incidence of injury in Iraq can be ascribed to the degradation of infrastructure and living standards endured by the populace, coupled with the lack of conventional hygienic facilities in specific areas. Furthermore, the existence of fauna and arthropods in highly concentrated and contaminated regions acts as a sustaining element for harm [22].

There is a paucity of research on the molecular identification of scabies in Iraq, with the exception of a study conducted by Naz et al. [23] in Pakistan, which is among the few studies on this topic worldwide. The present study conducted an analysis of the Polymerase Chain Reaction (PCR) technique on Sarcoptesscabiei mites that were obtained from patients suffering from scabies. The study employed a set of two primers for the purpose of the analysis. The band of 178 bpwas observed in lane 1 when Sarms 15 F/R was subjected to 2% agarose gel electrophoresis. Upon conducting the analysis using primer 16S D1/D2 on agarose gel, it was observed that lane 2 exhibited bands measuring 460 bp and 600 bp.

Wong et al. [24] conducted additional research in Hong Kong utilising the polymerase chain reaction (PCR) method to molecularly identify patients with scabies. Ninety-eight percent nucleotide identity was found between the S. scabiei type hominis cox1 gene and the PCR product sequenced from the 29 skin scrapings that tested positive for cox1-PCR.

The present study aims to investigate the feasibility of utilising Polymerase Chain Reaction (PCR) technique on skin scraping samples obtained from patients who are suspected to have scabies. Bae et al. [25] conducted a PCR analysis on skin scrapings in South Korea, with the aim of targeting the cytochrome c oxidase subunit 1 (cox1) gene of Sarcoptesscabiei.

The pathological findings presented in this study align with prior research indicating that the epidermis exhibits thickness and keratosis, accompanied by infiltration of inflammatory cells [26-27]. In reaction to the parasite, cellular immune mononuclear cells go to the site of infection. Female parasites dig tunnels in the dermis of the host

to deposit their eggs, causing inflammation and scarring. During burrowing, the female organism secretes chemicals that help penetrate the skin. The aforementioned materials function as antigenic molecules, thereby inducing the migration of inflammatory cells towards the infected regions, resulting in a heightened presence of immune and inflammatory cells in the affected areas.

The pathogenic consequences brought on by the presence of the infectious agent and its persistent secretions, as well as the immune system responses, are exacerbated by the chronicity of the infection[28].

Conclusion:

The findings of the present investigation indicate that scabies remains a neglected public health concern, with a significant number of individuals afflicted by this condition, resulting in various histological alterations and harm to the integumentary system.

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