DISTRIBUTION AND ANTIBIOTIC SENSITIVITY PROFILE OF SKIN INFECTION CAUSING PATHOGENS IN DISTRICT PESHAWAR, PAKISTAN

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ABSTRACT

OBJECTIVES

The study aimed to evaluate the distribution and antibiotic sensitivity profile of dermatophytes fungi and skin infection-causing bacterial pathogens in the district of Peshawar, Pakistan.

METHODOLOGY

A cross-sectional study was conducted from February 2022 to July 2022 in Microbiology Section, Complex Medical Laboratory Peshawar, Pakistan. A total of 100 skin-infected patients pus, nail, and skin scraping samples were processed for the isolation of fungal and bacterial pathogens.

RESULTS

Out of 100 skin-infected patient samples, the distribution of Escherichia coli was higher at 44.23%, followed by Staphylococcus aureus at 25%, Proteus species at 21.15%, Klebsiella spp. 5.76%, and Pseudomonas aeruginosa 3.84%, respectively. Among fungal pathogens, the distribution of Candida spp. was higher at 44.44%, followed by Aspergillus spp. 22.22%, Rhizopus spp. 16.16%, Mucor spp. 11.11%, Paecilomyces lilacinus 5.55%, respectively. The E. coli showed high resistance to amoxicillin 86.95%, S. aureus was high resistance to ciprofloxacin, levofloxacin 84.61%, Klebsiella spp. was found high resistance to amoxicillin and meropenem 100%, Proteus spp. has found high resistance to ciprofloxacin, amoxicillin 81.81%, and P. aeruginosa was highly resistant to doxycycline, aztreonam 100%. The candida spp. was found high resistance to nystatin at 87%, Aspergillus spp. were founded highly resistant to nystatin at 100%, Mucor spp. was high resistance to fluconazole, ketoconazole, and clotrimazole (100%), Rhizopus spp. was found resistant to itraconazole 100%, P. lilacinus was found highly resistant to itraconazole, nystatin 100%.

CONCLUSION

The study of antibiotic resistance pattern is suggested, which help the basis for modifications in skin infection therapy. A molecular study was also needed to identify the resistance gene among these pathogens and their immunogenicity.

KEYWORDS: *Dermatophytes, Skin infection, Antibiotic sensitivity, Peshawar, Pakistan*

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INTRODUCTION

Skin diseases are one of the most important segments of worldwide diseases, from which millions of people are infected.[1,2] According to the dermatologist, dermatophytes usually cause an infection of the skin.[3] One of the most common infectious disorders worldwide is dermatophytosis which creates many problems for public health. Dermatophytosis causes significant morbidity and however rises chiefly in an advanced state.[4]

Dermatology is a significant field where emergencies and mortality rarely occur. But in some conditions that do not provide proper treatment, this turns into extensive and severe lethal results.[5] In the medical department, dermatology is one of the most important branches which makes most of the diagnoses with visual observation. In standard care hospitals, there are many medical departments where dermatology departments are an integral part of standard care.[6] Dermatophytes are fungi that cause skin, nail, and hair infections. Dermatophytes are filamentous and superficial fungi. Dermatophytes commonly used host keratinized tissues to grow and can share and digest. Different species of dermatophytes cause mostly topical cutaneous fungal infection which creates infectious diseases.[7] In humans and animals, dermatophytes create mostly topical infections. Dermatophytes are considered highly pathogenic agents. Within the host, these micro-organisms are highly infectious, divide and multiply within the host keratinized tissues or organs for example, hair, nail, epidermal stratum corneum, skin, etc.[8] Dermatophytic fungi mostly belong to three genera: Trichophyton, Microsporum, and Epidermophyton.[9,10,11,12] Skin infections are not only caused by a fungus, but also many other types of bacteria that can cause skin infection in humans during surgical procedures, trauma, or post-burn injuries. Due to this type of infection pus is produced in the form of white to yellow fluid, which causes the death of white blood cells (WBC)**.** Both aerobic and anaerobic bacteria caused human skin infection, among them gram-positive bacteria include S. aureus and *S.* epidermidis, and gram-negative bacteria include E. coli, Acinetobacter spp., Citrobacter spp., Enterobacter spp., Pseudomonas spp., klebsiella spp., respectively.[13,14,15] The distribution of these pathogens strongly affects geographical locations. Therefore, the present study was designed to investigate the prevalence, and antibiotic sensitivity profile of dermatophytes fungi and skin infection-causing bacterial pathogens in the district of Peshawar, Pakistan.

METHODOLOGY

A cross-sectional study was conducted from February 2022 to July 2022 in the Microbiology section, of Complex Medical Laboratory Peshawar, Pakistan. A probability sampling technique was used in which a total of 100 skin-infected patients were recruited. For all infected patients, the informed consent form was signed. From skin-infected patient's pus, nail, and skin scraping samples were collected and transported to Microbiology, Complex Medical Laboratory Peshawar, Pakistan. For the isolation of bacterial pathogens Blood agar, MacConkey agar, and Chocolate agar were used and incubated at 37^{o}C for 24 hours. For the isolation of fungal pathogens, Sabourad dextrose agar was used and incubated at 25^{o}C for 02-03 days.[12] All positive samples or in which pathogen's growth was obtained are included in this study. All negative samples or in which no growth was obtained, are excluded from the study. The bacterial pathogens were identified based on colony morphology, gram staining, and biochemical test including the catalase test, coagulase test, oxidase test, urease test, citrate test, indole test, H_2S test, and TSI test. The fungal pathogens were identified based on colony morphology and microscopy. For microscopy, the fungal hyphae were stained by lactophenol stain. Furthermore, the urease test, Dermatophyte Test Media (DTM), and Corn Meal Agar (CMA) were also used for the identification of fungal pathogens. For bacterial and fungal antibiotic sensitivity testing the Clinical laboratory standard institution (CLSI, 2021) guidelines were followed. All obtained data was arranged and analyzed by statistical packages for social sciences (SPSS) 23.0 version software and Microsoft excel.

RESULTS

Out of 100 skin-infected patient samples, 70 samples were found positive while in 30 samples no growth was observed.

Table 1: Biochemical Characteristics of Bacterial Pathogens

Pathogens	Gram Stain	Indole Test	Oxidase Test	Catalase Test	Citrate Test	Urease Test
E. Coli	Negative	Positive	Negative	Positive	Negative	Negative
S. Aureus	Positive	Negative	Negative	Positive	Positive	Positive
Proteus Spp.	Negative	Positive	Negative	Positive	Positive	Positive
Klebsiella Spp.	Negative	Negative	Negative	Positive	Positive	Positive
P. Aeruginosa	Negative	Negative	Positive	Positive	Positive	Negative

Table 2: Occurrence of Skin Infection Causing Bacterial Pathogens

S. No.	Bacterial Pathogens	Number (%)
1.	E. coli	23 (44.23%)
2.	S. aureus.	13 (25%)
3.	Proteus spp.	11 (21.15%)
4.	Klebsiella spp.	03 (5.76%)
5.	P. aeruginosa.	02 (3.84%)

Table 3:Antibiotics Sensitivity and Resistance Pattern of Skin-Causing Bacteria

S. No.	Antibiotics		E. coli (n=13)	S. aureus (n=13)	Klebsiella spp. (n=3)	Proteus spp. (n=11)	P. aeruginosa (n=2)
1.	Amikacin (AK) 30µg	R	03 (13.04%)	02 (15.38%)	02 (66.66%)	02 (18.18%)	0 (0%)
		S	20 (86.95%)	11 (84.61%)	01 (33.33%)	09 (81.81%)	02 (100%)
2.	Meropenem (MEM) 10µg	R	07 (30.43%)	02 (15.38%)	03 (100%)	0 (0%)	0 (0%)
		S	16 (69.56%)	11 (84.61%)	0 (0%)	11 (100%)	02 (100%)
3.	Gentamicin (CN) 10µg	R	09 (39.13%)	03 (23.07%)	01 (33.33%)	03 (27.27%)	0 (0%)
		S	14 (60.86%)	10 (76.92%)	02 (66.66%)	08 (72.72%)	02 (100%)
4.	Ciprofloxacin (CIP) 5µg	R	18 (78.26%)	11 (84.61%)	01 (33.33%)	09 (81.81%)	0 (0%)
		S	05 (21.73%)	02 (15.38%)	02 (66.66%)	02 (18.18%)	02 (100%)
5.	Cefotaxime (CTX) 30µg	R	15 (65.21%)	01 (7.69%)	0 (0%)	03 (27.27%)	0 (0%)
		S	08 (34.78%)	12 (92.3%)	03 (100%)	08 (72.72%)	02 (100%)
6.	Levofloxacin (LEV) 5µg	R	18 (78.26%)	11 (84.61%)	02 (66.66%)	06 (54.54%)	01 (50%)
		S	05 (21.73%)	02 (15.38%)	01 (33.33%)	05 (45.45%)	01 (50%)
7.	Ampicillin (AMP) 10µg	R	17 (73.91%)	09 (69.23%)	02 (66.66%)	08 (72.72%)	0 (0%)
		S	06 (26.08%)	04 (30.76%)	01 (33.33%)	03 (27.27%)	02 (100%)
8.	Amoxicillin (AMC) 30µg	R	20 (86.95%)	05 (38.46%)	03 (100%)	09 (81.81%)	01 (50%)
		S	03 (13.04%)	08 (61.53%)	0 (0%)	02 (18.18%)	01 (50%)
9.	Doxycycline (DXT) 30µg	R	13 (56.52%)	01 (7.69%)	0 (0%)	10 (90.9%)	02 (100%)
		S	10 (43.47%)	12 (92.3%)	03 (100%)	01 (9.09%)	0 (0%)
10.	Aztreonam (ATM) 30µg	R	16 (69.56%)	06 (46.15%)	0 (0%)	03 (27.27%)	02 (100%)
		S	07 (30.43%)	07 (53.84%)	03 (100%)	08 (72.72%)	0 (0%)
11.	Ceftriaxone (CRO) 30µg	R	13 (56.52%)	09 (69.23%)	01 (33.33%)	02 (18.18%)	0 (0%)
		S	10 (43.47%)	04 (30.76%)	02 (66.66%)	09 (81.81%)	02 (100%)

Table 4: Morphological Characteristics of Isolated Fungal Pathogens

Pathogens	Colony Morphology	Microscopy
Rhizopus spp.	White, gray, fluffy growth reverse is gray or brown	Long, rare septa with dark terminal end
Mucor spp.	White, gray, fluffy growth reverse is white	Long, broad non-septate branched with round sporangia.
Candida spp.	White, smooth and yeast like appearance	Small, oval, unicellular, single budding may be seen
Aspergillus spp.	Blue-green with suede-like surface	Smooth and colorless spores which appears like plant
P. lilacinus	Fast growing, powdery or suede like gold, green, brown	Unicellular and form chain that can be intertangled

Table 5: Occurrence of Skin Infection Causing Fungal Pathogens

S. No.	Isolated Fungal Pathogen	Number (%)
1.	Candida spp.	08 (44.44%)
2.	Aspergillus spp.	04 (22.22%)
3.	Rhizopus spp.	03 (16.16%)
4.	Mucor spp.	02 (11.11%)
5.	P. lilacinus	01 (5.55%)

Table 6: Antifungal Sensitivity and Resistance Pattern of Skin-Causing Fungi

S. No.	Antifungal		Candida Spp. (N=8)	Aspergillus Spp. (N=4)	Mucor Spp. (N=2)	Rhizopus Spp. (N=3)	P. Lilacinus (N=1)
1.	Ketoconazole (Kt) 10µg	R	06 (75%)	03 (75%)	02 (100%)	02 (66.66%)	0 (0%)
		S	02 (25%)	01 (25%)	0 (0%)	01 (33.33%)	01 (100%)
2.	Clotrimazole (Cc) 10µg	R	05 (62.5%)	02 (50%)	02 (100%)	01 (33.33%)	0 (0%)
		S	03 (37.5%)	02 (50%)	0 (0%)	02 (66.66%)	01 (100%)
3.	Itraconazole (It) 10µg	R	04 (50%)	01 (25%)	01 (50%)	03 (100%)	01 (100%)
		S	04 (50%)	03 (75%)	01 (50%)	0 (0%)	0 (0%)
4.	Nystatin (Ns) 100u	R	07 (87.5%)	04 (100%)	0 (0%)	0 (0%)	01 (100%)
		S	01 (12.5%)	0 (0%)	02 (100%)	03 (100%)	0 (0%)
5.	Fluconazole (Flc) 10µg	R	0 (0%)	01 (25%)	02 (100%)	02 (66.66%)	0 (0%)
		S	08 (100%)	03 (75%)	0 (0%)	01 (33.33%)	01 (100%)

DISCUSSION

Dermatophytosis is an infection, which is caused by a group of fungi called keratophakic fungi. There are some favorable factors involved that promote fungal infection i.e., mycosis in the population that is poor hygiene environments, longtime contact with the household animals, poor social-economic, and overpopulation.[16] Besides these skin infections also caused by bacteria includes Micrococcus spp., S. aureus, E. coli., Klebsiella spp., Acinetobacter spp., Enterobacter, and Streptomyces on human skin.[16,17] Atopic dermatitis is a high-risk skin infection in patients, which has impressive causes of morbidity, but it may also develop systemic if left untreated. Commonly on the skin of the patient S. aureus colonizes with atopic dermatitis and is considered the most common micro-organisms to cause infections in the host.[18] In this regard, the current study was based on the evaluation of the prevalence, and antibiotic sensitivity profile of dermatophytes fungi, and skin infection-causing bacterial pathogens in the district of Peshawar, Pakistan. In the current study we found that out of 100 skin-infected patient samples, 70 samples were found positive, while out of 70 positive samples, bacterial growth was observed in 52 samples. The occurrence of E. coli was higher (44.23%), followed by S. aureus (25%), Proteus spp. (21.15%), Klebsiella spp. (5.76%), P. aeruginosa (3.84%), respectively. Our finding agrees with the previous studies of Khan et al. (2021), who reported that E. coli (46%) bacteria observed highly prevalent pathogens among the other bacterial pathogens followed by S aureus (39%), Proteus spp. (11%), Klebseilla spp. (2%), P. aerugenosa (2%).[12] According to another study findings Microsporum spp., Trychophyton spp., Epidermophyton spp., and Aspergillus spp., were highly prevalent dermatophytic pathogens in skin, hair, and nail samples.[13] In another study Trichophyton mentagrophytes, Trichophyton rubrum, Trichophyton violaceum, Epidermophyton floccosum, Microsporum gypseum, Trichophyton tonsurans, Trichophyton schoenleinii, and Trichophyton verrucosum were found mostly in skin fungal infections.[17] In agreement with these results, the current study showed that the occurrence of Candida spp. was higher (44.44%) as compared to other fungal pathogens followed by Aspergillus spp. (22.22%), Rhizopus spp. (16.16%), Mucor spp. (11.11%), P. lilacinus (5.55%), respectively. According to Khan et al. (2021) findings, E. coli was sensitive to Amikacin (90%) and highly resistant to Ampicillin (92.5%). S aureus reported being highly sensitive to Meropenem and Doxycycline (92.1%) and highly resistant to Levofloxacin (91.1%). Proteus spp. was highly sensitive to Meropenem (100%) and high resistance to Doxycycline (90%). Klebsiella spp. reported high sensitivity to Ciprofloxacin, Cefotaxime, Aztreonam and Doxycycline (100%) and showed resistance to Meropenem and Amoxicillin (100%). P. aeruginosa was highly sensitive to Amikacin, Meropenem, Ciprofloxacin, Gentamicin, Cefotaxime, Ceftriaxone, Ampicillin and Cefotaxime (100%) and resistant to Aztreonam and Doxycycline (100%).[12] In support of the previous studies our study, the sensitivity pattern of bacteria showed that E. coli notified resistance to Amoxicillin (86.95%) while highly sensitive to Amikacin (86.95%). S. aureus was high resistance to Ciprofloxacin, Levofloxacin (84.61%) and highly sensitive to Doxycycline, and Cefotaxime (92.3%). Klebsiella spp. was found high resistance to Amoxicillin and Meropenem (100%) while highly sensitive to Cefotaxime, Doxycycline and Aztreonam (100%). Proteus spp. has found high resistance to Ciprofloxacin, and Amoxicillin (81.81%) and highly sensitive to Meropenem (100%). P. aeruginosa was highly resistant to Doxycycline, Aztreonam (100%) while highly sensitive to Cefotaxime, Meropenem, Amikacin, Ampicillin, Ceftriaxone, Gentamicin, and Ciprofloxacin (100%). In our study sensitivity

pattern of fungi was found that candida spp. highly found resistance to Nystatin (87%), and highly sensitive to fluconazole (100%). Aspergillus spp. were founded high resistant to Nystatin (100%) and highly sensitive to itraconazole and fluconazole (75%). Mucor spp. was high resistance to fluconazole, ketoconazole and Clotrimazole (100%) while highly sensitive to Nystatin (100%). Rhizopus spp. was found resistant to itraconazole (100%) and highly sensitive to nystatin (100%). P. lilacinus was found high resistant to itraconazole, nystatin (100%), and highly sensitive to ketoconazole, clotrimazole and fluconazole. These findings agree with the previous study as they stated that six antifungal drugs are most commonly used to treat dermatophytosis: fluconazole, miconazole, clotrimazole, ketoconazole, griseofulvin, and terbinafine.[18]

LIMITATIONS

The limitation of this study was small and a molecular study was also needed to identify the resistance gene among these pathogens and their immunogenicity.

CONCLUSIONS

The E. coli and S. aureus was found most prevalent. The Candida spp., Aspergillus spp., Rhizopus spp., Mucor spp., and P. lilacinus were found fungal pathogens responsible for skin infection. The Candida spp. was found most prevalent. The bacterial pathogen's sensitivity pattern highly notified resistance to amoxicillin, ciprofloxacin, levofloxacin, Meropenem, doxycycline, and Aztreonam. The fungal pathogens sensitivity pattern highly notified resistance to nystatin, fluconazole, ketoconazole, clotrimazole, and itraconazole.

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