



Histopathological Pattern of Fungal Infections Seen in Usmanu Danfodiyo University Teaching Hospital Sokoto from 2014 To 2018

M. O. Mohammed¹, M. Sirajo¹, O. O. Okechi², A. T. Muhammad¹, R. I. Tsamiya¹, U. Abubakar^{1*}, I. Mohammed¹, A. Umar¹, M. K. Dallatu³, A. S. Ajayi³, S. M. Sani¹, A. Abdulazeez³, A. A. Hali¹, M. Isah¹, S. Y. Ma'aruf¹, B. A. Bello³, J. M. Bunza³, H. Kabir¹, F. A. Dogondaji¹, H. M. Tambuwal¹, M. Sirajo¹, N. Okorie⁴, A. A. Ngaski⁵, D. Isah⁶, S. M. Sani¹, A Salisu¹, N. A. Idris⁷, A. Ya'u¹, H. I. Wasagu¹ and H. Abdullahi⁸

¹Department of Histopathology, School of Medical Laboratory Science, Usmanu Danfodiyo University Sokoto, Nigeria.

²Department of Medical Laboratory Science, Abia State University, Uturu, Nigeria.

³Department of Histopathology, Usmanu Danfodiyo University Teaching Hospital Sokoto, Nigeria.

⁴Department of Medical Laboratory Science, Faculty of Health Sciences, Ebonyi State University, Nigeria.

⁵Department of Chemical Pathology, School of Medical Laboratory Science, Usmanu Dandofiyo University, Sokoto, Nigeria.

⁶Department of Haematology, School of Medical Laboratory Science, Usmanu Danfodiyo University, Sokoto, Nigeria.

⁷Department of Medical Laboratory Science, General Hospital Maiyama, Maiyama Local Government, Kebbi State, Nigeria.

⁸Department of Immunology, School of Medical Laboratory Science, Usmanu Danfodiyo University, Sokoto, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Author MOM owner of the work and perform the design of the work. Authors AAH, MI and SYM carried out and source the materials for literatures review. Authors OOO and MKD overall cross the work to ensure that everything is in order. Authors AU, RIT, UA and HK Carried out photomicrographs and interpretation of the results. Authors ATM and IM overall reviewer of the work. Authors NO, HMT and MS managed references. Authors AAN, JMB and DI carried out preparation of the stains used. Authors BAB, HA and FAD take care of the tissue processing. Authors SMS, ASA, AA, HIW and NAI managed the staining procedures and carried out screening of the slides.

Article Information

DOI: 10.9734/AJRID/2020/v4i330151

Editor(s):

(1) Dr. Gonzalo Emiliano Aranda Abreu, Veracruzana University, Mexico.

(2) Dr. Giuseppe Murdaca, University of Genoa, Italy.

Reviewers:

(1) Tayo Julius Bogunjoko, Eye Foundation Hospital Group, Nigeria.

(2) Adib SAAD, Tishreen University, Syria.

Complete Peer review History: <http://www.sdiarticle4.com/review-history/55913>

ABSTRACT

Introduction: Fungal infection is a common manifestation in this part of the country and worldwide. It is essential to define the epidemiology of fungal infection in a particular environment.

Aim: This study aims to analyze the diagnosis of fungal infection carried out in patients attending the histopathology department (UDUTH) Sokoto from 2014 to 2018.

Method: This study is a retrospective study of all fungal infection biopsies carried out from 2014 – 2019 in Usmanu Danfodiyo University Teaching Hospital, Sokoto, northwestern Nigeria, a referral centre for the neighbouring northern states Kebbi and Zamfara. Ethical clearance was obtained from the ethical committee of UDUTH, data was collected from histological report cards and analyze manually. Some tissue blocks of the case were retrieved to confirm the diagnosis. A total of 66 requests for fungal diagnosis were received from January 2014 to December 2018, which represent about 0.7% of the total biopsies received over the study period.

Result: Out of the 66 of request for fungal diagnosis 32(48.5%) were fungal positive. Subcutaneous mycosis was the most frequent fungal infection with a frequency of 18(56.25%). Males were more frequently affected than females with the frequencies of 18(56.25%) and 14 (43.75%) respectively.

Keywords: Fungal infection; epidemiology; diagnosis; Nigeria.

1. INTRODUCTION

Fungi are prokaryotic to the eukaryotic organism. They are neither plant nor animal, they are also not bacteria. When all the organisms are categorised as either plant or animal, fungi were categorised as plants. Plants produce nutrients through photosynthesis and animal-derived nutrient through eating these plants and other animals. On the other hand, fungi derive nutrients in other ways such as through degradation of both plant and animal remains [1,2]. Fungal infections are caused by a variety of fungi with a variety of manifestation. The illness caused by fungi can range from superficial [athlete's foot] to deadly [blood stream infection]. The three general classes of fungal infection that the mycotic disease branch of the centres for disease control is concern about are opportunistic infection, community-acquired infections, and hospital-acquired infections [3,4]. Fungal infections are also classified into superficial mycosis, cutaneous mycosis, subcutaneous mycosis, and systemic mycosis [5,2]. The incidence of fungal infection is increasing at an alarming rate presenting an enormous challenge to health care professionals. This increase is directly related to the growing population of immunocompromised individuals resulting from the change in medical practice such as the use of intensive chemotherapy and immunosuppressive drugs. HIV and another

disease which causes immune suppression have also contributed to this problem. The superficial and subcutaneous fungal infection affects skin, keratinous tissue and mucous membrane. Including in this class are some of the most frequently occurring skin disease affecting millions of people worldwide. However, they are rarely life-threatening. The systemic fungal infection may be caused by either opportunistic organism that infects an at-risk host or may be associated with the more invasive organism that is endemic to the specific area systemic fungal infection can be life-threatening and are associate with high morbidity and mortality [6,7]. Cutaneous, subcutaneous and systemic fungal infections are those that affect body tissue. Most of the cutaneous and subcutaneous infections are easily diagnose and readily amiable to treatment. systemic fungal infection can be life-threatening and are associated with high morbidity and mortality [6,8]. Invasive fungal infections are generally distinguished from superficial mycosis based on the involvement of other sterile body sites or invasion of organ tissue. Although superficial mycosis accounts for most of the overall global prevalence of fungal infection, invasive fungal infections are associated with disproportional high morbidity or mortality and economic burden [9,10]. Cutaneous and subcutaneous mycosis is a source of significant morbidity in both immunocompetent and immunocompromised

patients. A growing number of fungal infections are often associated with disseminated disease. Skin and soft tissue biopsy allow mycological identification [11,7]. Histopathological examination of tissue detected fungal invasion of tissue and vessels as well as the host reaction to the fungus, it is and will remain an important tool to define the diagnostic significance of positive culture isolate or result from PCR testing [6,12]. Isolated soft tissue infections are uncommon but may cause severe morbidity or mortality among transplant recipients and other immunosuppressed patients [7,13]. Cutaneous and subcutaneous mycosis is an evolving field. Clinician all over the world should be aware of this common manifestation of this disease – infectious disease as they increasingly reported and may lead to or associated with dissemination [14,15,11,5]. A total of 3374 invasive fungal infection episode occur in 3154 patients. The mean incidence was 27.29 (100 000 patients) per year. Although the invasive fungal infection rate varies from year to year ($r^2 = 0.9$), linear regression estimated a mean amount increase of 0.24\100 000 or 0.9% per year ($p = 21$). *Candida* spp were most common (55.2) dimorphic primary *coccidiosis* spp comprised 25.5% followed by *Aspergillus* spp (0-97) year 13.4% of cases occurred in children less than 18 years. Comorbidities were common include diabetes mellitus (28.7%) and chronic pulmonary disease (44.9%). active malignancy was present in 13.2% autoimmune or primary immunodeficiency in 14.9% and 5.9% of the episode occurred in transplant recipients 26.1% of invasive fungal infection occurred inpatient receiving immunosuppressive therapy of which corticosteroid was the most common (20.8% of all invasive fungal infection) lymphopenia (absolute lymphocyte count less than 500 cell\mm³) was present at the time of invasive fungal infection in 22.1% of the episode. Hospital admission occurred in 76.2%; invasive fungal infection occurred during intensive care unit stay in 30.7% of cases [16,17,18]. In Nigeria, estimate indicates that over 11.8% of the Nigerian population is estimated to suffer from serious fungal infection each year. If *Tinea capitis* and recurrent vaginal thrush are excluded over 960 000 are estimated to be affected with substantial mortality [19,9,20,21].

2. MATERIALS AND METHODS

2.1 Study Area

This study was carried out in Usmanu Danfodiyo University Teaching Hospital UDUTH Sokoto state, Nigeria.

2.2 Study Population

This involved histologically confirmed case of fungal infection from 2014 to 2018.

2.3 Study Design

This study is a retrospective study. Data was collected retrospectively over five years from 2014 to 2018.

2.4 Inclusion Criteria

Demographic data such as age, gender, and histopathological diagnosis was collected from the histological record of the subjects.

2.5 Data Collection

Data was collected from histology record book and histopathological report cards of the positive subjects. The total number of biopsies received over the study period was counted; the number of requests for fungal diagnosis was also noted. Some previous blocks were also retrieved.

2.6 Data Analysis

Data was analysed by using statistical package for social science SPSS version 20.

2.7 Results

Results were expressed in the statistical table, bar charts and pie charts.

2.8 Methodology

Sample: Some of the representative formalin-fixed, paraffin-embedded tissue blocks of the cases were used. Thin sections were cut and fresh Grocott's modification of Gomori's methenamine silver (GMS) stained section was produce.

2.9 Procedure for Grocott'S Modification of Gomori'S Methenamine Silver

1. The section was taken into the water
2. The section was oxidized in 5% chromic acid for 1 hour.
3. The section was rinsed in water
4. The section was rinsed in 1% sodium bisulphite to remove residual chromic acid
5. The section was rinsed in water
6. The section was washed in 3 changes of distilled water

7. The section was taken into working methenamine solution pre-heated at 60°C until the section turns yellowish-brown
8. The section was washed in 6 changes of distilled water
9. The section was toned in 0.1% gold chloride for 5 minutes
10. The section was rinsed in distilled water
11. The section was treated with 2% sodium thiosulphate for 5 minutes to removed unreduced silver
12. The section was washed in water
13. The section was counterstained with light green for 30 seconds
14. The section dehydrated, cleared and mounted with DPX [22]

Controls: Already stained fungal positive section was used as a positive control and fungal negative section was stained and used as the negative control.

3. RESULTS

A total of 66 requests for fungal diagnosis were received from January 2014 to December 2018, which represent about 0.7% of the total biopsies received over the study period. Out of the 66 number of request for fungal diagnosis 32(48.5%) were fungal positive. Subcutaneous mycosis was the most frequent fungal infection with the frequency of 18 (56.25) followed by systemic mycosis with the frequency of 12(37.50%). See Table 1.

Out of the 32 cases, 18(56.25%) were males and 14 (43.75%) were females. See Table 2.

The most frequently age group affected is the age between 21 to 30 years with the Frequency of 10 (28.25%), then followed by age group between 31 to 40 years with the frequency of 9 (28.13%). The rest are age group between 1 to 10 years, 11 to 20 years, 41 to 50 years, 51 to 60 years with the frequency of 3 (9.38%), 4 (12.50%), 3 (9.38%) and 3 (9.38%) respectively. See Table 3.

Out of 2 cutaneous mycoses, 0 (0%) were male and 2 (100%) were female, in subcutaneous mycosis 11 (61.1%) subjects were male while 7 (38.9%) were female and in systemic mycosis 7 (58.3%) were male while 5 (41.7%) were female. See Tables 4, 5 and 6.

Out of 2 subjects with cutaneous mycosis 1 (50%) is in the group between 21 to 30 years while the other 1 (50%) is in the age group between 51 and 60 years. See Table 7.

The most frequent age group with subcutaneous mycosis is the age between 31 to 40 years with the frequency of 5 (27.78%). See Table 8.

The most frequent age group with systemic mycosis is the age between 21 to 30 years with the frequency of 5 (41.67%). See Table 9.

Table 1. Frequency and percentage (%) distribution of fungal infection

Types of mycosis	Frequency	Percentage (%)
Superficial mycosis	0	00
Cutaneous mycosis	2	6.25
Subcutaneous mycosis	18	56.25
Systemic mycosis	12	37.50
Total	32	100

Table 2. Frequency and percentage (%) distribution of mycosis in relation to gender

Gender	Frequency	Percentage (%)
Male	18	56.25
Female	14	43.75
Total	32	100

4. DISCUSSION

A total of 66 number of request for fungal diagnosis were received from January 2014 to December 2018. This is about 0.7% of the total number of biopsies received over the studyperiod. out of the 66 number of requests 32 (48.5%) were fungal positive. Subcutaneous mycosis was the most frequent mycosis, this does not agree with the study by Lakshmanan et al., [12;23;24]. which showed that superficial fungal infection is the most common in tropical and sub-tropical countries. However, most of the superficial mycosis are diagnosed in the microbiology department. Also according to [15; 25;7]. Cutaneous, subcutaneous and systemic fungal infection are those that affect body tissue. [17;26;27], show that subcutaneous mycosis second most frequently mycosis after superficial mycosis, this can be related to this study.

Table 3. Frequency and percentage (%) distribution of mycosis in relation to age group

Age group (years)	Frequency	Percentage (%)
1-10	3	9.38
11-20	4	12.50
21-30	10	31.25
31-40	9	28.13
41-50	3	9.38
51-60	3	9.38
Total	32	100

Table 4. Frequency and percentage (%) distribution of cutaneous mycosis in relation to gender

Gender	Frequency	Percentage (%)
Male	0	00
Female	2	100
Total	2	100

Table 5. Frequency and percentage (%) distribution sub-cutaneous mycosis in relation to gender

Gender	Frequency	Percentage (%)
Male	11	61.1
Female	7	38.9
Total	18	100

In this study out of the 32 number of fungal infection 18 (56.25%) were male while 14 (43.75) were female, this is in contrast with the study made by Cheng *et al.* [16;28], which show that out of 518 patients with fungal infection 382 (73.7%) were males while 136 (26.3%) were females. In this study the age range is between 0 to 60 years and the most frequent age group with fungal infection is the age between 21 to 30 years with the frequency of 10 (31.25%) followed by age between 9 (28.13%). About famine gender, this can be contrasted with the study made by [5;29;1]. Which show that the prevalence among the age group in female was age between 20 to 29 years, however in the male the prevalent age group is the age between 60 to 69 years.

The distribution of different mycosis *i.e.*, cutaneous, subcutaneous and systemic mycosis in relation to gender are 0 (0%) male while 2

(100%) female, 11 (61.1%) male while 7 (38.9%) female and 7 (58.3%) male while 5 (41.7%) female respectively. Male has a high prevalence of subcutaneous mycosis, [8;30], shows that males are more prone to sub-cutaneous injury.

Table 6. Frequency and percentage (%) distribution of systemic mycosis in relation to gender

Gender	Frequency	Percentage (%)
Male	7	58.3
Female	5	41.7
Total	12	100

Table 7. Frequency and percentage (%) distribution of cutaneous mycosis in relation to age groups

Age group (years)	Frequency	Percentage (%)
1-10	0	0,0
11-20	0	0.0
21-30	0	0.0
31-40	1	50.0
41-50	0	0.0
51-60	1	50.0
Total	2	100

Table 8. Frequency and percentage (%) distribution of subcutaneous mycosis in relation to age groups

Age group (years)	Frequency	Percentage (%)
1-10	3	16.87
11-20	2	11.11
21-30	4	22.22
31-40	5	27.78
41-50	2	11.11
51-60	2	11.11
Total	18	100

Table 9. Frequency and percentage (%) distribution of systemic mycosis in relation to age groups

Age group (years)	Frequency	Percentage (%)
1-10	0	0.0
11-20	2	16.67
21-30	5	41.67
31-40	4	33.33
41-50	1	8.33
51-60	0	0.0
Total	12	100

Table 10. Frequency prevalence of fungal infection based on year

Types of mycosis	Years (frequency)					Total
	2014	2015	2016	2017	2018	
Superficial mycosis	0	0	0	0	0	0
Cutaneous mycosis	0	0	1	0	1	2
Subcutaneous mycosis	2	7	4	3	2	18
Systemic mycosis	0	3	1	3	6	12
Total	2	10	6	8	9	32

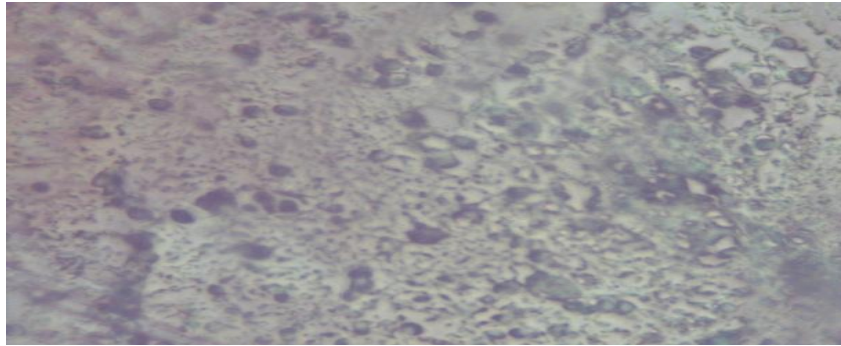


Plate 1. Photomicrograph showing chronic fungal rhinosinusitis Grocott's methenamine silver staining technique X400

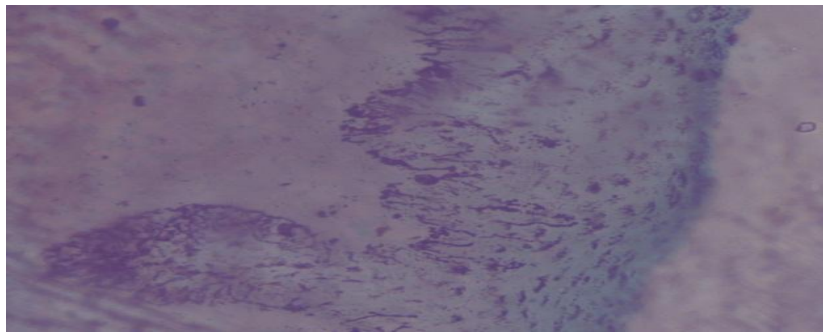


Plate 2. Photomicrograph showing actinomycosis Grocott's methenamine silver staining technique X400

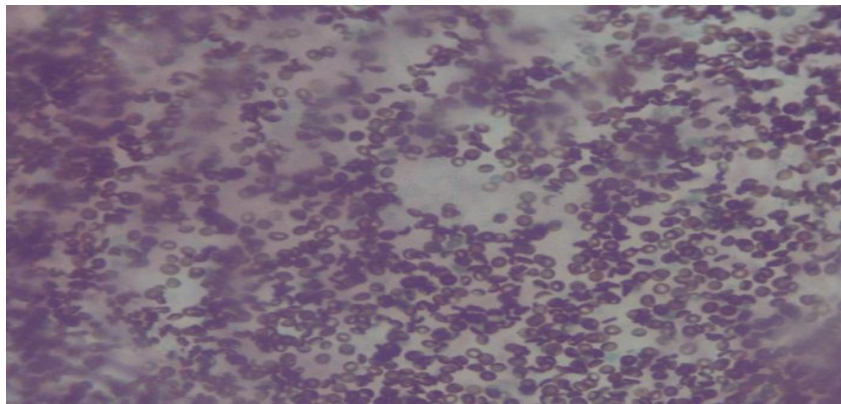


Plate 3. Photomicrograph showing deep mycosis (Blastomyces) Grocott's methenamine silver staining technique X400

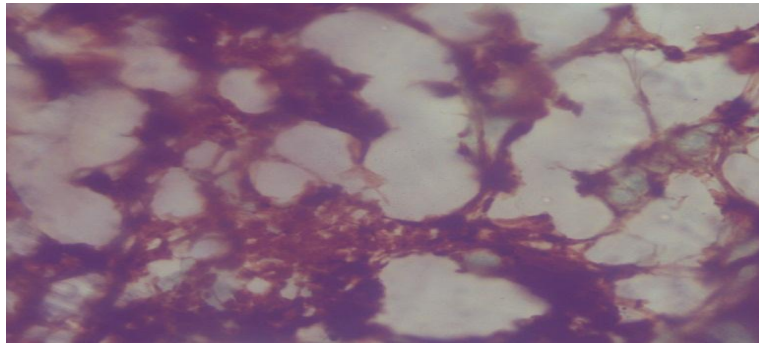


Plate 4. Photomicrograph showing fungal granulomatous inflammation Grocott's methenamine silver staining technique X400

The prevalent age group with subcutaneous mycosis is the age between 31 to 40 years with the frequency of 5 (27.78%) while the age group between 21 to 30 years has a higher prevalence of systemic mycosis.

5. CONCLUSION

This study was carried out to analyse the fungal diagnosis in histopathology laboratory Usmanu Danfodiyo University Teaching Hospital and to determine the most frequent type of fungal infection and occurrence about gender and age group.

There was more subcutaneous mycosis. Males were more frequently affected than females. The most frequent age group is the age between 21 to 30 years then followed by age between 31 to 40 years hence, age between 21 to 40 years was the most frequent age group affected with fungal infection.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Ethical approval was obtained from the ethical committee of Usmanu Danfodiyo University Teaching Hospital, Sokoto.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Lakshmanan A, Ganeshkumar P, Raam SM, Hemamalina M, Madhavan R. Epidemiological and clinical pattern of dermatomycoses in rural India. *Indian journal of medical microbiology*. 2015;33: 134-136.
2. Warren L. Review of medical microbiology and immunology 14th edition. Lange Clinical use of oral nystatin in the prevention of systemic candidosis in patients at particular risk. *J. Mycoses*. 2014;39:329-339.
3. CDC Fungal disease. Terbinafine: a review of its pharmacodynamics and pharmacokinetic properties, and therapeutic potential in superficial mycoses. 2012;43:259-284
4. Darouiche RO. Candida in the ICU. *Clinical Chest Medicine*. 30:287-293.
5. Joshi P, Yadav R, Singh G. Histological identification of Entomophthoromycosis in biopsy samples are required. *Indian Journal of Pathology and Microbiology*. 2014;57:514-516.
6. Guarner J, Brandt ME. Histopathologic diagnosis of fungal infections in the 21st century. *Clinical Microbiology Rev*. 2011;24:247-280.
7. Heinz T. Soft tissue fungal infection: surgical management of 12 immunocompromised patients. *Plastreconst surg*. 1996. Huston SM, Mody CH. Cryptococcosis: an emerging respiratory mycosis. *Clinical Chest Medicine*. 2009;30:253-264.
8. Pradhan SV, Talwar OP, Ghosh A, Swami RM, Shiva Raj KC, Gupta S Chromoblastomycosis in Nepal: A study of 13 cases. *Indian Journal Dermatol Venereol Leprol*. 2007;73(2):176-8.
9. Parish J.M, Blair JE. Coccidioidomycosis. *Mayo Clinical Procedure*. 2008;83:343-349.

10. Ribes JA, Vanover-Sams CL, Baker DJ. Zygomycetes in human disease. *Clinical Microbiology Revised*. 2000;13:236-301.
11. Guagen S, Fungal skin and soft tissue infection. *Curropin infect Dis*. 2006;12(2):31-34.
12. Lin TY, Yeh KM, Lin JC, Wang NC, Peng MY, Chang FY. Cryptococcal disease in patients with or without human immunodeficiency virus: clinical presentation and monitoring of serum cryptococcal antigen titers. *Journal Microbio Immunol Infection*. 2009;42:220-226.
13. Oladele RO. Burden of serious fungal infection in Nigeria. *West African journal Medicine*, 2014;12(3):45-46.
14. Alberti C, Bouakline A, Ribaud P Relationship between environmental fungal contamination and the incidence of invasive aspergillosis in haematology patients. *Journal of Hospital Infection*. 2001;48(3):198-206.
15. Gazzoni AF, Severo CB, Salles EF, Severo LC Histopathology, serology and cultures in the diagnosis of cryptococcosis. *Rev Inst Med Trop Sao Paulo*. 2009;51:255-259.
16. Cheng S, Chong L. A prospective epidemiological study on tinea pedis and onychomycosis in Hong Kong. *Chinese Medical Journal*. 2002;115(6):860-865.
17. Concia E, Azzini AM, Conti M. Epidemiology, incidence and risk factors for invasive candidiasis in high-risk patients. *Journal of Drugs*. 2009;69(1):5-14.
18. Queiroz-Telles F, Esterre P, Perez-Blanco M, Vitale RG, Salgado CG, Bonifaz A overview of clinical manifestations, diagnosis and treatment. *Medical Mycology*. 2009;47:3-15.
19. Fidel PLJ. History and update on host defense against vaginal candidiasis. *American Journal Reprod Immunology*. 2007;57(5):2-12.
20. Sayal SK, Prasad GK, Jawed KZ, Sanghi S, Satyanarayana S. Chromoblastomycosis. *Indian J Dermatol Venereol Lepro*. 2002.;68:233-234.
21. Surendran K, Bhat RM, Bolor R, Nandakishore B, Sukumar D. A clinical and mycological study of dermatophytic infections. *Indian J Dermatol*. 2014;59:262-7. US Gov.
22. Awriro OG, Principle and technique of histochemistry and tissue pathology 5th edition. Clavarianum press Ibadan; 2014.
23. Roden MM, Zaoutis TE, Buchanan WL, et al. Kontoyiannis DP, Walsh TJ. Epidemiology and outcome of zygomycosis: a review of 929 reported cases. *Clin Infect Dis*. 2005;41:634.
24. Sargent J, O'Marcaigh A, Smith O, Butler K, Gavin P, O'Sullivan M. *Candida albicans* associated necrotizing vasculitis producing life-threatening gastrointestinal hemorrhage. *Hum. Pathol*. 2010;41:602-609.
25. Gupta N, Arora SK, Rajwansi A, Nijhawan R, Srinivasan R. Histoplasmosis: cytodiagnosis and review of literature with special emphasis on differential diagnosis on cytomorphology. *Journal of Cytopathology*, 2010;21:240-244.
26. Hee JY, Hwa YC., Young KK., Yeong JS, Moran K. Prevalence of fungal infection using national health insurance data from 2009-2013, South Korea. *Epi H*, 2013;36: 4-7.
27. Sullivan DC, Chapman SW. Bacteria that masquerade as fungi: *Actinomycosis/nocardia*. *Proc Am Thorac Soc*. 2010;7:216-221.
28. Dwari BC, Ghosh A, Paudel R, Kishore PA. Clinicoepidemiological study of 50 cases of cutaneous tuberculosis in a tertiary care teaching hospital in Pokhara, Nepal. *Indian Journal Dermatol*. 2009;55: 233-237.
29. Jyunichi PA, Shigeki I, Takamichi O, Ami S, Kozue S. Introduction to the world of fungi. *Pub mycology society of japan*; 2016.
30. Riscili BP, Wood KL. Noninvasive pulmonary *Aspergillus* infections. *Clinical Chest Medicine*. 2009;30:315-335.

© 2020 Mohammed et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:
 The peer review history for this paper can be accessed here:
<http://www.sdiarticle4.com/review-history/55913>