

## CUTANEOUS ASPERGILLOSIS AND CANDIDIASIS ASSOCIATED WITH RICE FARMING IN PARTS OF EBONYI STATE, NIGERIA

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### Abstract

Cutaneous aspergillosis and candidiasis associated with agricultural practices in Nigeria have a major health impact on rice farmers and other associated people. This leads to reduced productivity through decreased human resources, famine and places an additional burden on health care requirements, hence, the study on the distribution of cutaneous aspergillosis and candidiasis among rice farmers in Ebonyi State. A total of 2130 rice farmers were randomly selected and screened for cutaneous fungal infections. Hair fragments, skin and nail scrapings were collected using sterile scissors and scalpels from 182 rice farmers that presented with lesions suggestive of the infection. The samples were examined by direct microscopy using 5% potassium hydroxide (KOH) and cultured on Sabouraud dextrose agar supplemented with 0.05mg/ml chloramphenicol and incubated at 28°C ± 2°C for 2 days. The fungal isolates were identified by macroscopic and microscopic methods, and confirmed by molecular analysis. Demographic data and factors influencing the infection were obtained through questionnaires and analyzed using analysis of variance (ANOVA) at  $p < 0.05$  significant level with SPSS version 22. A total of 143 fungal isolates were recovered and included *Aspergillus flavus* 64 (44.76%), *A. tamarii* 41 (28.67%), *A. nomius* 30 (20.98%) and *Candida albicans* 8 (5.59%). Age, gender, educational status and family size of the farmers and their interactions with domestic animals/pets influenced the distribution of the fungal species. Out of the 182 rice farmers screened, 16-26 years age-group [71 (39.01%)] were most infected with the infection, followed by 27-37 years age-group [39 (21.43%)] and a decline between the age-group 49-59 years [15 (8.24%)]. The females were more infected 144 (79.12%) than the males 38 (20.88%). The infection among the females was found to be significant ( $P < 0.05$ ). In-depth knowledge of the etiologic agents of the infections and potential risk factors associated with rice farming in Ebonyi State could provide insights on control measures. In addition, effective reporting of the infections, risk awareness and appropriate intervention measures will assist in tackling the infections amongst rice farmers in Ebonyi State and the country at large.

**Keywords:** cutaneous aspergillosis, candidiasis, rice farmers, agricultural practices, health impact, intervention

## INTRODUCTION

Cutaneous aspergillosis is a skin disease caused by a variety of species of *Aspergillus* [1]. *Aspergillus* species are among the most ubiquitous fungi, seen in soil, water, decaying vegetations and any substrate that contains organic debris. The fungi enter the body through a break in the skin and causes infection, usually in people who have weakened immune systems. In recent years, fungal infections related to *Aspergillus* species have become a major focus of clinical microbiology and infectious disease studies as the number of patients infected with *Aspergillus* species has risen dramatically. Among the known *Aspergillus* species, *Aspergillus fumigatus* is the most common one causing infections in humans, whereas *Aspergillus flavus* is as important as *A. fumigatus* and is the second most common *Aspergillus* species associated with human infections [2].

Cutaneous candidiasis is a fungal infection of the skin and nail plates caused by members of the genus *Candida* and other related genera. *Candida* species are saprophytic, yeast-like fungi. The infection usually occurs in warm, moist and creased areas, such as the inguinal or intergluteal areas. It is a fairly common opportunistic disease and is usually caused by maceration and trauma to the skin and nails [3, 4]. Moisture, heat, friction, and maceration of the skin are the principle predisposing factors in the normal patient [5]. The clinical manifestations may be acute, subacute or chronic to episodic. They usually remain superficial and respond readily to treatment. *C. albicans*, *C. tropicalis*, *C. parapsilosis*, and *C. glabrata*, are responsible for 50–90% of all cases of candidiasis in humans [6, 7].

Rice farming is one of the major occupations of the people of Ebonyi State, Nigeria. This grain, which is one of the worlds' most important food crops, is cultivated either in marshy low land areas with plenty of water or in plateau or hilly regions where natural rainfall provides adequate amount of water. These farmers are believed to be at high risk of cutaneous fungal infections because of their occupational contact with soil. They are exposed to various irritants such as mud, cow dung or other types of manure, fertilizers, dust and marshy soil that predispose them to the infection [8, 9].

Cutaneous aspergillosis and candidiasis are highly contagious and represent a significant public health problem in Nigeria and the world at large [10]. Although not usually life-threatening, the infection can be persistent and symptomatic, with millions of Naira spent annually in their treatment [11]. Generally, the infections have been reported worldwide, though with variation in distribution, incidence, epidemiology, etiology and hosts from one location to another with time. Host susceptibility may be enhanced by moisture, warmth, specific skin chemistry, composition of sebum and perspiration, age, exposure and genetic predisposition [11]. Other factors such as personal hygiene, geographical location, climatic factors, social practices, health care, crowded living and poor sanitary conditions, socioeconomic status and immigration may influence the

prevalence of cutaneous fungal infections [12, 13]. The incidence is higher in hot humid climates and crowded living conditions.

A number of studies on the distribution and etiological aspects of cutaneous aspergillosis and candidiasis have been carried out in different parts of Nigeria which had school children as their target population [10, 14-17]. There is paucity of epidemiological data in the literature to ascertain the distribution of cutaneous aspergillosis and candidiasis among rice farmers in Ebonyi State. Thus, studies in different geographic areas assessing the specific fungal etiology involved are of public health importance serving as baseline information for the management of the infections at the local level.

## **MATERIALS AND METHODS**

### **Study Area**

Ebonyi State is located in the South Eastern part of Nigeria and is bounded to the North by Benue State, to the East by Cross River, to the South by Abia State and to the West by Enugu State. It is a tropical climatic zone. It has an average temperature of 28°C and humidity of 80–90% in dry season. The vegetation is guinea savanna characterized by a mixture of deciduous trees, shrubs, and grasses. Majority of the people in the study area are farmers who cultivate large quantities of rice, yam and cassava for sale. There are significant patches of swamp which form the basis of their agricultural activities. There are ponds, streams, springs and other stagnant water bodies scattered all over the area [18].

### **Collection of Samples**

Six communities including Okpera, Omega achara, Nkalafor, Agubia, Ndechi and Igbudu in Ikwo Local Government Area and Iboko, Ikeanyi, Amaguanyim, Sharon, Mgbo and Ezza Ohu in Izzi Local Government Area of Ebonyi State were chosen for the study. An informed consent was taken prior to the collection of samples. Samples were collected during field visits and house-to-house visit with the help of a guide who knew the local residents involved in rice field work. A total of 2130 rice farmers including adults and children were randomly chosen and interviewed.

The rice farmers were screened for lesions suggestive of cutaneous fungal infections on their skin, hair and nails. For obtaining the samples aseptically, the infected areas or lesions were cleaned with cotton wool soaked in 70% ethanol before collecting scrapings from the affected areas using sterile scalpels. The scrapings were put into drug dispensary envelopes and transported to the laboratory for examination. The examination was carried out within 24 hour. The information about the age, gender, location of lesion, educational level, size of family, interaction with domestic animals and applications of antifungal therapy were obtained through questionnaire.

### **Examination of Direct Potassium Hydroxide Mount**

Direct microscopic examination was carried out as described by [19]. A part of the hair follicles, scrapings of skin and nails were treated with two drops of a 5% potassium hydroxide (KOH) using a sterile forceps for 10 minutes to allow clearing of epithelial tissue surrounding the fungi so that the fungal structures can be exposed. The preparation was mounted on a clean glass slide and examined by direct microscopy for the presence of fungal hyphae and arthrospores under low power (X10) and high power (X40) magnification. The positive samples were processed for the isolation of fungal species on Sabouraud Dextrose Agar (SDA; Lab M).

### **Isolation of Dermatophytes**

Culture of samples was carried out as described by [19]. A portion of the sample was inoculated on the prepared culture media (SDA) supplemented with 0.05mg/ml chloramphenicol (Yangzhou, China) in duplicates using spot inoculation technique. The inoculated plates were incubated at room temperature ( $28^{\circ}\text{C} \pm 2^{\circ}\text{C}$ ) for 2 days. Where fungal growth was observed, the organism was subcultured onto fresh medium to obtain pure isolate used for identification.

### **Identification of Fungal Isolates**

Preliminary identification of the fungal isolates was carried out based on detailed phenotypic methods: colony morphology, growth rate, lactophenol cotton blue stain, slide culture and germination tube test. The morphologies were compared with the standard description given by [20]. Confirmatory identification of the fungal isolates was done by sequencing the internal transcribed spacer (ITS) region of the ribosomal DNA.

### **DNA Isolation**

DNA was extracted from fresh fungal cultures on Sabouraud agar medium. Colonies were picked up with sterilized toothpick, suspended in 100 $\mu\text{l}$  of sterilized saline in a PCR tube and centrifuged at 10,000 rpm for 10 minutes. After removal of supernatant, the pellet was suspended in 50 $\mu\text{l}$  of InstaGene Matrix (Bio-Rad, USA). Incubated at  $56^{\circ}\text{C}$  for 30 minutes and then heated at  $100^{\circ}\text{C}$  for 10 minutes.

### **PCR Amplification, Purification and DNA Sequencing**

Internal transcribed spacer 1 and 4 (ITS1 and ITS4) used for the PCR were designed as ITS1 forward primer 5' (TCC GTA GGT GAA CCT GCG G) 3' and ITS4 reverse primer 5' (TCCTCC GCT TAT TGA TAT GC) 3'. The PCR reaction was performed with 20 ng of genomic DNA as the template in a 30 $\mu\text{l}$  reaction mixture by using a *EF-Taq* (SolGent, Korea) as follows: activation of Taq polymerase at  $95^{\circ}\text{C}$  for 2 minutes, 35 cycles of  $95^{\circ}\text{C}$  for 1 minute,  $55^{\circ}\text{C}$ , and  $72^{\circ}\text{C}$  for 1 minute each was performed, finishing with a 10 minute step at  $72^{\circ}\text{C}$ .

The amplification products were purified with a multiscreen filter plate (Millipore Corp., Bedford, MA, USA). Sequencing reaction was performed using a PRISM Big Dye Terminator v3.1 Cycle sequencing Kit. The DNA samples containing the extension products were added to Hi-Di formamide (Applied Biosystems, Foster City, CA). The mixture was incubated at 95 °C for 5 minutes, followed by 5 minutes on ice and then analyzed by ABI Prism 3730XL DNA analyzer (Applied Biosystems, Foster City, CA).

### Statistical Analysis

The distribution of *Aspergillus* and *Candida* species among rice farmers in Ebonyi State was expressed in percentage. In order to establish the association of age, gender, educational level, size of family, interaction with domestic animals and location of lesion with the epidemiology of aspergillosis and candidiasis among the farmers, the analysis of variance (ANOVA) statistical test was used at  $p < 0.05$  significant level. Statistical computation was done using Statistical Package for Social Science (SPSS) version 22 for windows.

## RESULTS

A total of 2130 rice farmers were sampled from six communities each of Ikwo Local Government Area (Table 1) and Izzi Local Government Area of Ebonyi State (Table 2).

Table 1: Distribution of rice farmers screened for lesions suggestive of cutaneous fungal infections in Ikwo LGA of Ebonyi State

| Community       | Number sampled |
|-----------------|----------------|
| Number infected |                |
| Okpera          | 388            |
|                 | 41             |
| Omega achara    | 201            |
|                 | 10             |
| Nkalafor        | 274            |
|                 | 18             |

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|        |      |
|--------|------|
| Agubia | 90   |
|        | 10   |
| Ndechi | 41   |
|        | 7    |
| Igbudu | 150  |
|        | 26   |
| Total  | 1144 |
|        | 112  |

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Table 2: Distribution of rice farmers screened for lesions suggestive of cutaneous fungal infections in Izzi LGA of Ebonyi State

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| Community  | Number sampled | Number infected |
|------------|----------------|-----------------|
| <hr/>      |                |                 |
| Iboko      | 142            |                 |
|            | 12             |                 |
| Ikeanyi    | 297            |                 |
|            | 28             |                 |
| Amaguanyim | 180            |                 |
|            | 8              |                 |
| Sharon     | 118            |                 |
|            | 5              |                 |
| Mgbo       | 122            |                 |
|            | 7              |                 |

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|          |     |
|----------|-----|
| Ezza ohu | 127 |
|          | 10  |
| Total    | 986 |
|          | 70  |

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Out of the 2130 rice farmers screened, only 182 (8.54%) persons presented with lesions suggestive of cutaneous fungal infections and comprised of 22 (1.03%) males and 90 (4.23%) females in Ikwo LGA (Table 3) and 16 (0.75%) males and 54 (2.53%) females in Izzi LGA (Table 4). The tables showed that the females were more infected with cutaneous fungal infections than their male counterparts. The 16-26 years age group was the most infected in both Ikwo LGA 48 (2.25%) and Izzi LGA 23 (1.08%).

Table 3: Demographic characteristics of rice farmers that presented with lesions suggestive of cutaneous fungal infections in Ikwo LGA of Ebonyi State

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| Age Groups           | Gender |        | of        |
|----------------------|--------|--------|-----------|
| specimens<br>(Years) | Number |        |           |
|                      | Male   | Female | collected |
| 5-15                 | 1      | 9      | 10 (0.47) |
| 16-26                | 5      | 43     | 48 (2.25) |
| 27-37                | 10     | 11     | 21 (0.99) |
| 38-48                | 3      | 20     | 23 (1.08) |

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|        |           |            |
|--------|-----------|------------|
| 49-59  | 3         | 7          |
|        |           | 10 (0.47)  |
| Total  | 22 (1.03) | 90         |
| (4.23) |           | 112 (5.26) |

Table 4: Demographic characteristics of rice farmers that presented with lesions suggestive of cutaneous fungal infections in Izzi LGA of Ebonyi State

| Age Groups | Gender    |        | of            |
|------------|-----------|--------|---------------|
| samples    | Number    |        |               |
| (Years)    |           |        |               |
| (%)        | Male (%)  | Female | collected (%) |
| 5-15       | 1         | 8      | 9 (0.42)      |
| 16-26      | 2         | 21     | 23 (1.08)     |
| 27-37      | 7         | 11     | 18 (0.85)     |
| 38-48      | 5         | 10     | 15 (0.70)     |
| 49-59      | 1         | 4      | 5 (0.23)      |
| Total      | 16 (0.75) | 54     | 70 (3.28)     |
| (2.53)     |           |        |               |



Out of the 182 specimens collected, 136 were positive by KOH mount whereas all the specimens collected were positive by culture. The positive KOH mount showed fragments of septate and nonseptate hyphae, and budding yeast cells. A total of 143 fungal isolates were recovered from the specimens. The phenotypically identified fungal species were further confirmed by molecular identification using internal transcribed spacer (ITS) region. The ITS regions were successfully amplified and sequenced from all the fungal isolates by the fungus-specific universal primers ITS1 and ITS4 at Macrogen Inc, Amsterdam, Netherlands.

The prevalence rates of cutaneous aspergillosis and candidiasis in Ikwo and Izzi Local Government Areas of Ebonyi State were 5.26% (112/2130) and 3.28% (70/2130) respectively (Figure 1). The overall prevalence rate of cutaneous aspergillosis and candidiasis among rice farmers in Ebonyi State was 8.54% (182/2130).

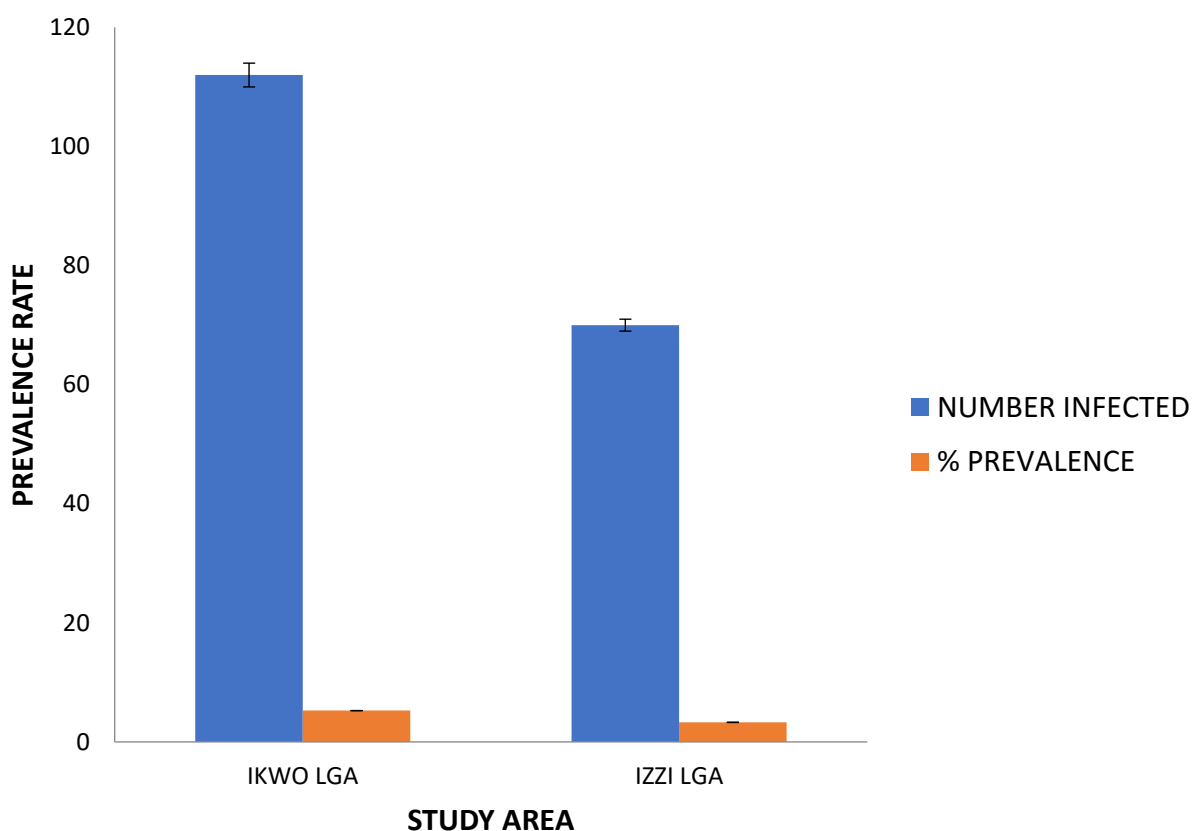


Figure 1: Prevalence rates of cutaneous aspergillosis and candidiasis among rice farmers in Ikwo and Izzi LGAs of Ebonyi State

*Aspergillus flavus* was the predominant species found in 34 (23.78%) cases of fungi isolated, followed by *A. tamarii* 15 (10.49%), *A. nomius* 15 (10.49%) and 5 (3.49%) cases of *Candida albicans* in Ikwo LGA (Figure 2). Figure 3 showed that *A. flavus* 30 (20.98%) was the predominant fungal species isolated, followed by *A. tamarii* 26 (18.18%), *A. nomius* 15 (10.49%) and *C. albicans* 3 (2.10%) in Izzi LGA.

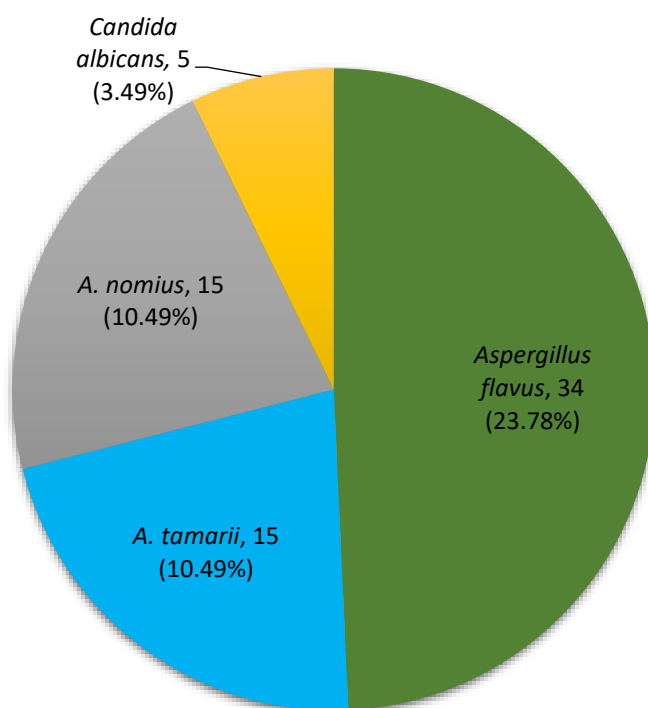


Figure 2: Percentage distribution of *Aspergillus* species and *Candida albicans* isolated from rice farmers in Ikwo LGA of Ebonyi State

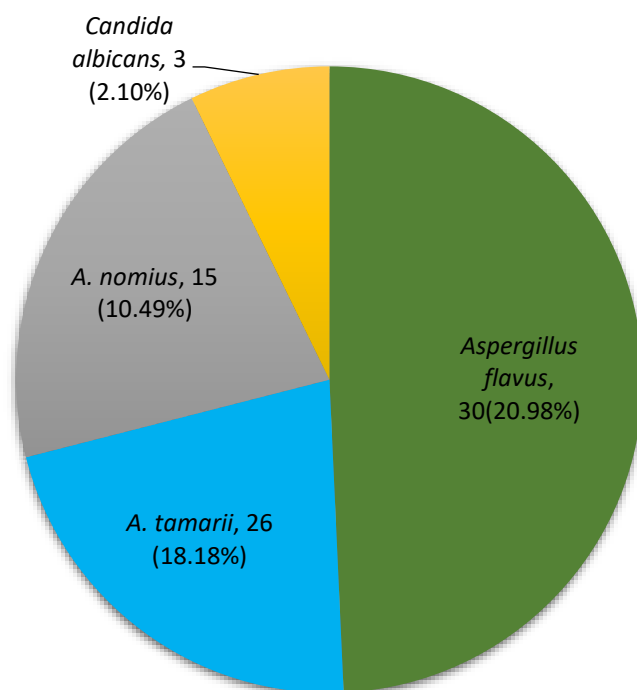


Figure 3: Percentage distribution of *Aspergillus* species and *Candida albicans* isolated from rice farmers in Izzi LGA of Ebonyi State

Out of the 143 fungal species identified in the study, 69 (48.25%) was isolated from Ikwo LGA. The highest percentage frequency 17 (11.89%) of fungal species was observed among rice farmers in Nkalafor community followed by Agubia 15 (10.49%), Okpera and Ndechi had 10 (6.99%) each while Igbudu and Omega achara had 9 (6.29%) and 8 (5.60%) respectively in Ikwo LGA. *A. flavus* 13 (6.60%), *A. nomius* 6 (3.04%) and *A. tamarii* 4 (2.03%) were the most frequently isolated species in Nkalafor, Agubia and Omega achara respectively (Figure 4).

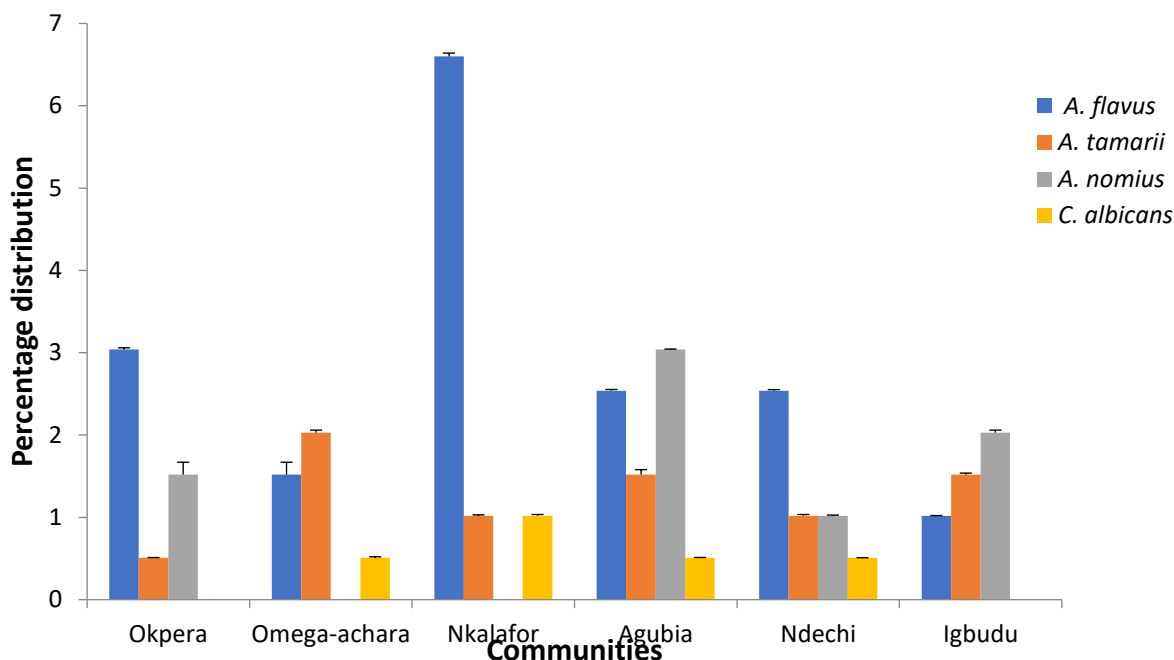


Figure 4: Percentage distribution of *Aspergillus* species and *Candida albicans* based on communities in Ikwo LGA of Ebonyi State

Of the total fungal species isolated, 74 (51.75%) was recovered from Izzi LGA. The highest percentage frequency 14 (9.79%) of fungal species was observed among rice farmers in Ikeanyi and Ezza ohu communities each, followed by 12 (8.39%) in Amaguanyim and Sharon each, and 11 (7.69%) in Iboko and Mgbo each. *A. flavus* 8 (4.06%), *A. tamarii* 6 (3.04%) and *A. nomius* 5 (2.54%) occurred mostly in Agbanyim, Sharon and Ikeanyi respectively (Figure 5).

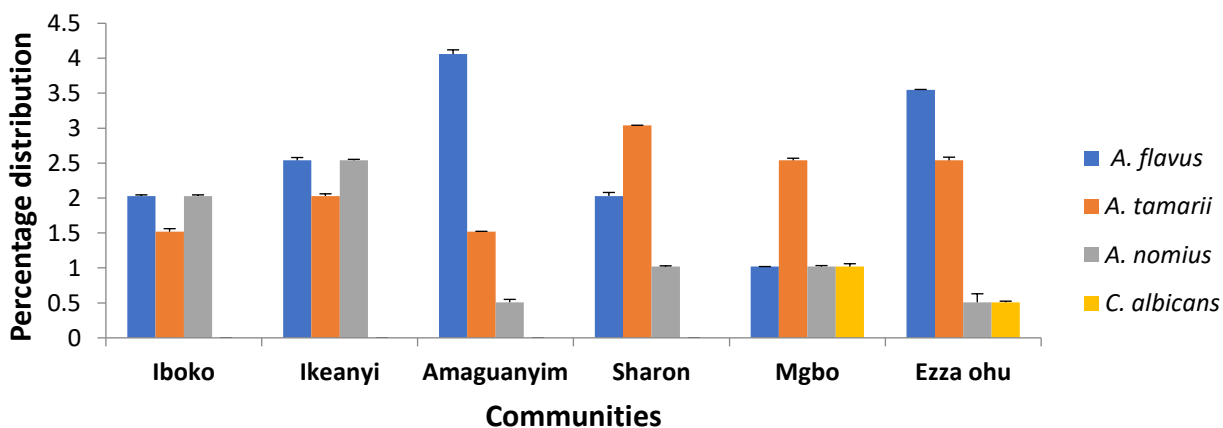


Figure 5: Percentage distribution of *Aspergillus* species and *Candida albicans* based on communities in Izzi LGA of Ebonyi State

The distribution of *Aspergillus* species and *Candida albicans* within age groups in Ikwo and Izzi LGAs showed that the infection occurred mostly among age-group 16-26 years 30 (15.24%), followed by 27-37 years 15 (7.61%) in Ikwo LGA whereas Izzi LGA recorded 16-26 years 27 (13.70%) and 27-37 years 19 (9.65). *A. flavus* caused infection in all age groups examined in the study. The age-group 16-26 years was most infected with *A. flavus* 17 (8.62%) and *A. tamarii* 10 (5.08%) in Ikwo LGA and Izzi LGA respectively (Figure 6). The age-group 5-15 years was most infected with *C. albicans* 3 (1.52%) in Ikwo LGA followed by 2 (1.02%) in Izzi LGA. Statistical analyses revealed significant difference ( $P < 0.05$ ) in the prevalence rates of cutaneous aspergillosis and candidiasis among rice farmers in all age groups.

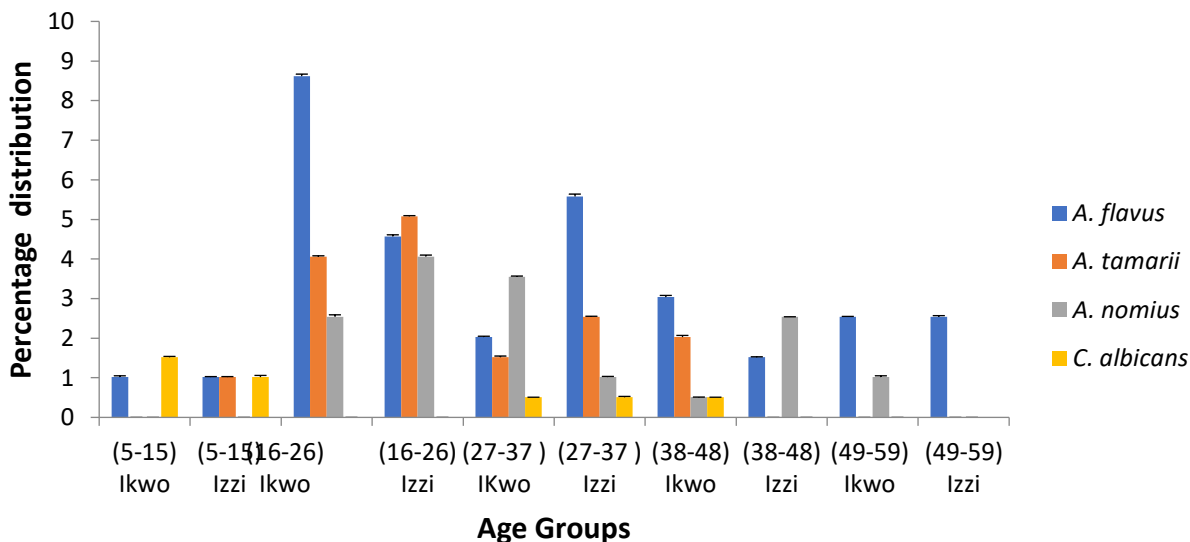


Figure 6: Percentage age distribution of *Aspergillus* species and *Candida albicans* among rice farmers in Ikwo and Izzi LGAs

The gender-specific distribution of *Aspergillus* species and *Candida albicans* among rice farmers in Ikwo LGA was 12 (6.09%) and 57 (28.94%) while in Izzi LGA was 16 (8.12%) and 58 (29.44%) for male and female farmers respectively. The females were more infected with aspergillosis and candidiasis in both LGAs. *A. flavus* 30 (15.23%) and *A. tamarii* 24 (12.18%) were the most frequently isolated species in Ikwo and Izzi LGAs respectively (Figure 7). Comparison of the prevalence rates of aspergillosis and candidiasis in males and females showed that female rice farmers had a significantly higher ( $P < 0.05$ ) prevalence rate.

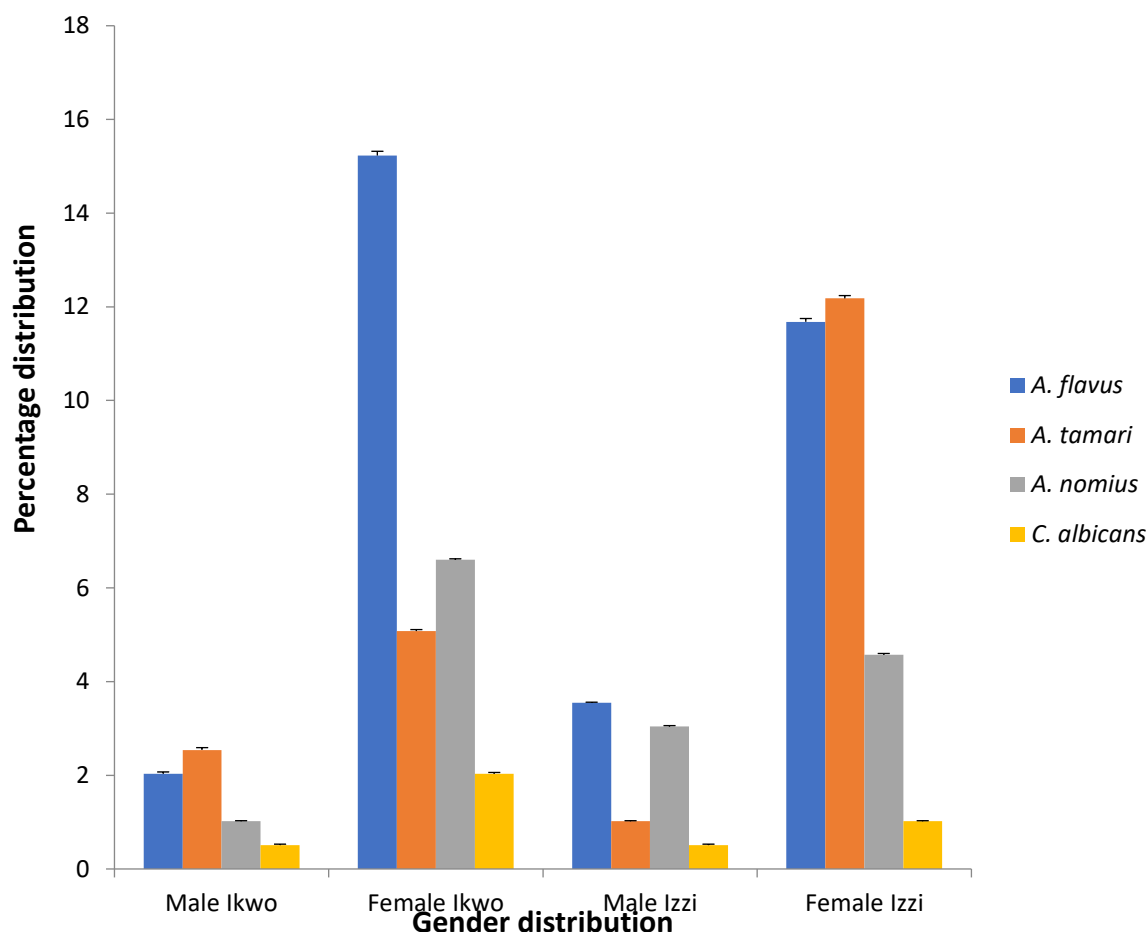


Figure 7: Percentage gender distribution of *Aspergillus* species and *Candida albicans* among rice farmers in Ikwo and Izzi LGAs of Ebonyi State

Highest frequencies 35 (15.23%) and 44 (15.23%) of aspergillosis and candidiasis were observed among rice farmers with no formal education in Ikwo and Izzi LGAs respectively. The prevalence was more in Izzi LGA. The least frequencies 4 (2.03%) and 2 (1.02%) of the infection were observed among those with tertiary education in Ikwo and Izzi LGAs respectively, followed by secondary education 15 (7.61%) and 12 (6.09%), and primary education 15 (7.61%) and 16 (8.13%) in Ikwo and Izzi LGAs respectively. *A. flavus* and *A. nomius* caused infection in all the educational groups examined in both Ikwo and Izzi LGAs (Figure 8). *A. flavus* was most frequently isolated in Izzi LGA 23 (11.68%) than Ikwo LGA 13 (6.60%).

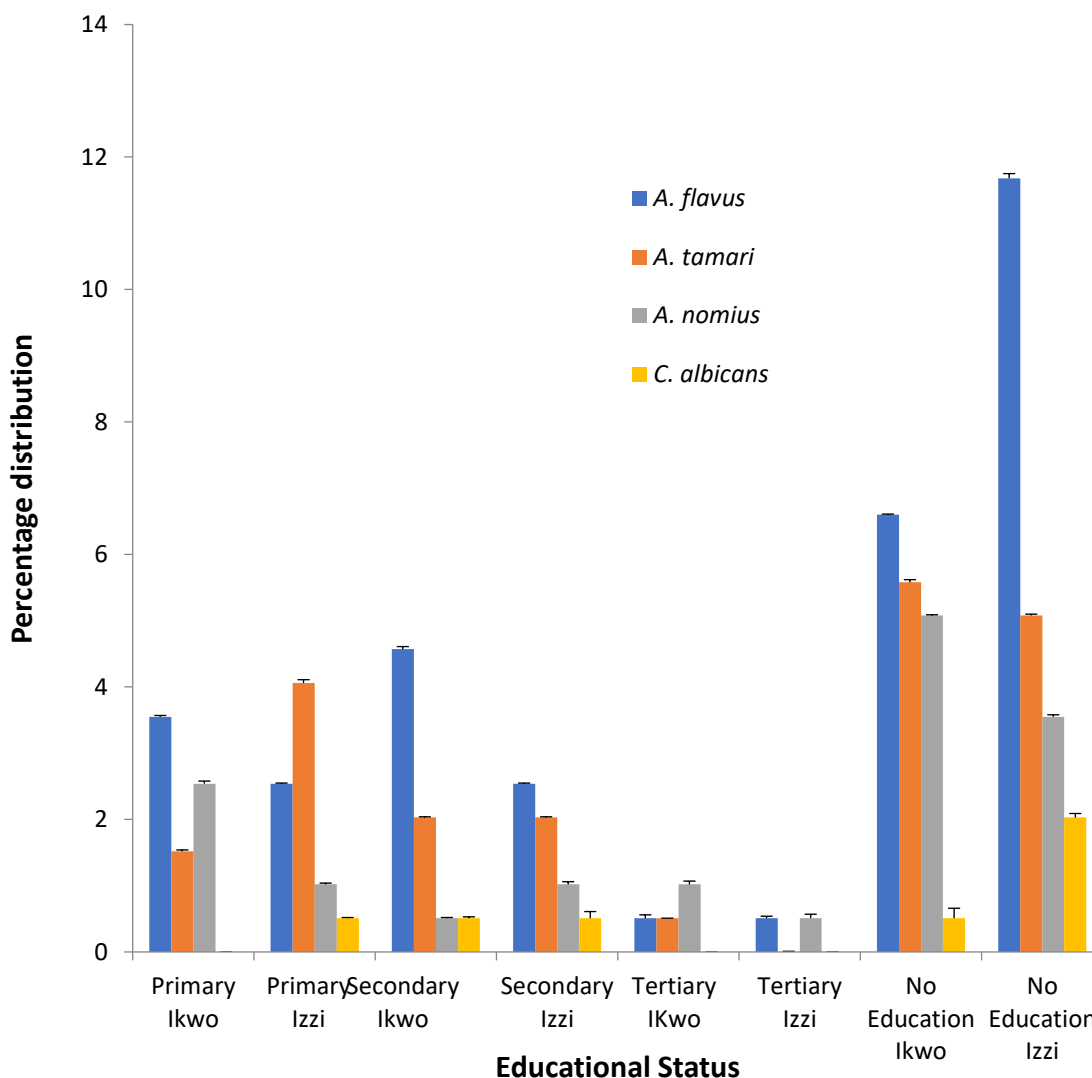


Figure 8: Percentage distribution of *Aspergillus* species and *Candida albicans* based on educational status of rice farmers in Ikwo and Izzi LGAs of Ebonyi State

In the distribution of aspergillosis and candidiasis based on family size of the rice farmers, highest percentage frequency 32 (16.24%) of *Aspergillus* species and *Candida albicans* was observed among rice farmers with families of 12-14 persons in Izzi LGA compared to 24 (12.18%) in Ikwo LGA, followed by 9-11 persons 26 (13.20%) and 18 (9.14%) in Ikwo and Izzi LGAs respectively. *A. flavus* and *A. nomius* were the most frequently isolated species found on all family sizes of rice farmers examined in both Ikwo and Izzi LGAs. *A. flavus* 13 (6.60%) occurred most among rice farmers with both family sizes of 9-11 and 12-14 persons in Ikwo and Izzi LGAs respectively (Figure 9). Infection due to *C. albicans* was not observed among rice farmers with families of 3-5 persons in both Ikwo and Izzi LGAs.

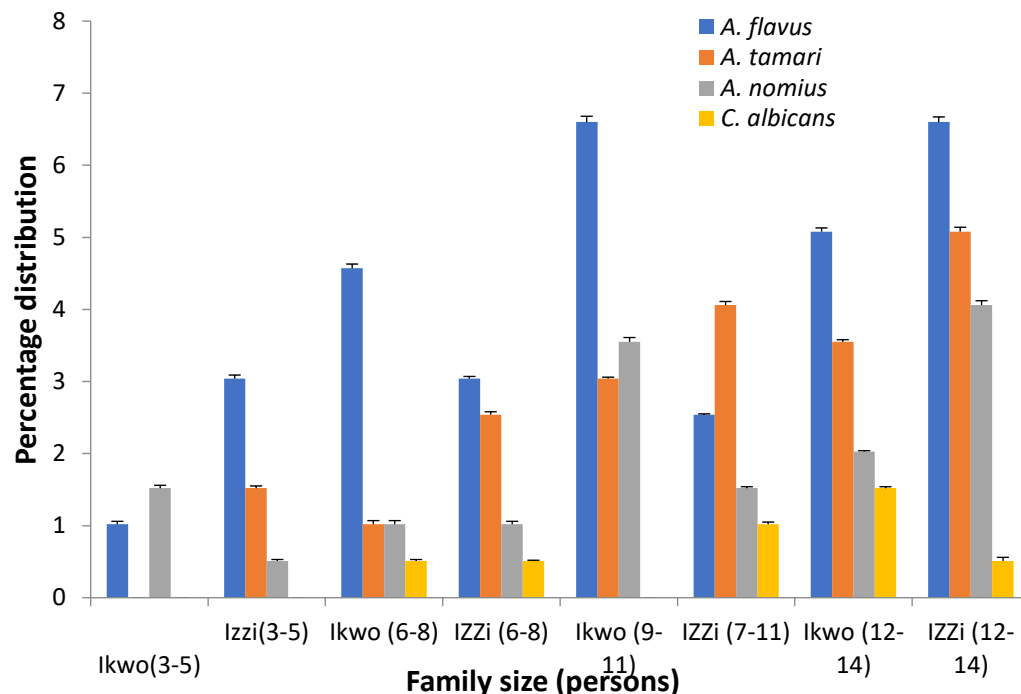


Figure 9: Percentage distribution of *Aspergillus* species and *Candida albicans* based on family size of rice farmers in Ikwo and Izzi LGAs of Ebonyi State

The infection was also reported among those that interact with pets/ domestic animals such as dogs, cats, goats, rabbits and poultry birds. Highest percentage frequencies 69 (35.03%) and 74 (37.56%) of *Aspergillus* species and *Candida albicans* were observed among rice farmers with no interaction with pets/ domestic animals in Ikwo LGA (Table 5) and Izzi LGA (Table 6) respectively.

Table 5: Percentage distribution of *Aspergillus* species and *Candida albicans* among rice farmers in Ikwo LGA based on their interactions with domestic animals

| Domestic Animals  | Fungal species | No. of Rice Farmers | No. of Isolates (%) |
|-------------------|----------------|---------------------|---------------------|
| Goats             |                |                     | A.                  |
| <i>flavus</i> (2) |                |                     |                     |
|                   |                | 2                   | 2                   |
| (1.02)            |                |                     |                     |
| Poultry           |                |                     | A.                  |
| <i>flavus</i> (3) |                |                     |                     |



|                      |                       |                    |
|----------------------|-----------------------|--------------------|
|                      | 5                     | 5                  |
| (2.54)               |                       |                    |
| <i>nomius</i> (2)    |                       | <i>A.</i>          |
| Cats                 |                       | <i>A.</i>          |
| <i>flavus</i> (1)    |                       |                    |
|                      | 3                     | 3                  |
| (1.52)               |                       |                    |
| <i>nomius</i> (2)    |                       | <i>A.</i>          |
| No animal            |                       | <i>A. flavus</i>   |
| (21)                 |                       | 59                 |
| 59 (29.95)           |                       |                    |
| interaction          |                       | <i>A. tamaraii</i> |
| (17)                 |                       |                    |
| (soil and wet works) | <i>A. nomius</i> (18) |                    |
|                      |                       | <i>C.</i>          |
| <i>albicans</i> (3)  |                       |                    |
| Total                |                       |                    |
|                      |                       | 69 (35.03)         |

Table 6: Percentage distribution of *Aspergillus* species and *Candida albicans* among rice farmers in Izzi LGA based on their interactions with domestic animals

| Domestic Animals    | Fungal species | No. of Rice Farmers | No. of Isolates (%) |
|---------------------|----------------|---------------------|---------------------|
| Goats               |                |                     | <i>A.</i>           |
| <i>flavus</i> (1)   |                |                     |                     |
|                     |                | 1                   | 1                   |
| (0.51)              |                |                     |                     |
| Poultry             |                |                     | <i>A.</i>           |
| <i>flavus</i> (1)   |                |                     |                     |
|                     |                | 3                   | 3                   |
| (1.52)              |                |                     |                     |
|                     |                |                     | <i>A.</i>           |
| <i>tamaraii</i> (2) |                |                     |                     |

|                      |          |                      |                 |
|----------------------|----------|----------------------|-----------------|
| Dogs                 |          |                      | A.              |
| <i>nomius</i> (1)    |          | 1                    |                 |
|                      | 1 (0.51) |                      |                 |
| Cats                 |          |                      | A.              |
| <i>flavus</i> (1)    |          |                      |                 |
|                      | 1        | 1                    |                 |
| (0.51)               |          |                      |                 |
| No animal            |          | A.                   | <i>flavus</i>   |
| (34)                 |          |                      | 46              |
| 68 (34.51)           |          |                      |                 |
| interaction          |          | A.                   | <i>tamaraii</i> |
| (22)                 |          |                      |                 |
| (soil and wet works) |          | A. <i>nomius</i> (7) |                 |
|                      |          |                      | C.              |
| <i>albicans</i> (5)  |          |                      |                 |
| Total                |          |                      |                 |
|                      |          | 74 (37.56)           |                 |

The locations of cutaneous lesions and the organisms recovered are presented in Table 7. The table showed that the scalp 76 (3.57%) was the commonest affected site of the body among the suspected cases. This was followed by the nail 59 (2.77%) and the skin 47 (2.20%). *A. flavus* was recovered from both hair and nail lesions examined in the study.

Table 7: Distribution of cutaneous aspergillosis and candidiasis according to body sites

| Body site affected<br>(%)          | No. sampled<br>Organisms |
|------------------------------------|--------------------------|
| recovered                          |                          |
| Hair                               | 76 (3.57)                |
|                                    | A. <i>flavus</i> , A.    |
| <i>tamaraii</i> , A. <i>nomius</i> |                          |

|                            |                         |
|----------------------------|-------------------------|
| Nail                       | 59 (2.77)               |
| <i>nomius, C. albicans</i> | <i>A. flavus, A.</i>    |
| Skin                       | 47 (2.20)               |
| <i>albicans</i>            | <i>A. tamaritii, C.</i> |
| Total                      | 182 (8.54)              |

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## DISCUSSION

Cutaneous aspergillosis and candidiasis constitute an important public health problem among rice farmers worldwide, including Nigeria. This disease remains endemic in Nigeria, largely because of lack of information on its prevalence and absence of control measures [10]. The present study highlights the distribution of cutaneous aspergillosis and candidiasis among rice farmers in Ebonyi State, Nigeria. Ikwo and Izzi, the two Local Government Area chosen for the study have relatively higher population density consisting primarily of rice farmers. In this study, there were more females than males due to the fact that rice farming in the study areas was done mostly by females.

The study revealed that 182 (8.54%) of rice farmers in a random population of 2130 farmers were infected with various species of cutaneous *Aspergillus* and *Candida albicans*. The prevalence of cutaneous aspergillosis and candidiasis observed in the study was insignificant and was unexpected because almost all parameters known to favour person to person transmission, such as the presence of skin lesions, practice of sharing personal belongings, overcrowding in the home, practice of keeping domestic animals/pets and factors that suggest unhygienic life style of the community with low socio-economic background are present in the rice farmers surveyed. Moreover, rice farming is generally associated with higher risk of developing cutaneous mycotic infection, both due to contact with many potential sources of infection such as animals, soil and wet work which promote infection [8, 9, 12, 21]. Similar observations were reported from a study among rice farmers in Anambra State [22]. The low prevalence may be due to variation in

environmental and climatic conditions favourable for the growth of the pathogens in the area studied. Another possible explanation for the low prevalence of cutaneous aspergillosis and candidiasis observed in this study is that the study only looked at those with detectable signs of fungal infection. This has the potential of missing asymptomatic carriers.

*A. flavus* was the predominant species followed by *A. tamaraii* and *A. nomius*. It can be deduced that *A. flavus* was the major aetiologic agent of aspergillosis among rice farmers in the study area. The finding of *A. flavus* as the predominant species was at variance with similar research by [22]. However, because no such study had been previously carried out in the study area among the studied population, the assertion of a variation in etiology is possible. The frequency of *Aspergillus* species among the sampled population of rice farmers was significant compared to *Candida albicans* ( $P < 0.05$ ). The high frequency in the isolation of *Aspergillus* species as observed in this study would suggest that those unhygienic practices that establish aspergillosis specifically are still much present within the area surveyed. A high frequency in isolation of non-dermatophyte molds was also recorded by [8, 23] and was attributed to their regular isolation from water, air, soil and vegetation. This would have to be dealt with in order to achieve reduction in the load of infection with fungal organisms generally as rice farming poses high chances of their getting in contact with human skin, giving rise to opportunistic infection.

Although rice farmers of all ages were susceptible to cutaneous aspergillosis and candidiasis, most of them belong to the age group 16-26 years. Similar observation was made by [22]. The age group 16-30 years is more actively involved in rice farming and exposed to the hot and humid climate than the younger and older age groups and tends to sweat more. Gender-related studies on the prevalence of cutaneous aspergillosis and candidiasis consist of fragments, with some studies claiming that males predominate [24, 25] while in some, it is the females [10, 26, 27]. This study also found more females than males infected with cutaneous aspergillosis and candidiasis. The probable reason for the higher prevalence of the infection amongst females in the study could be because there were more females in the areas surveyed than males as well as more females that presented with the infection, thus this may have made the gender prevalence of the males to be lower than that of the females.

The present study showed a relationship between the level of education and the prevalence of cutaneous aspergillosis and candidiasis in the population studied. Lower frequencies of species of cutaneous *Aspergillus* and *Candida albicans* were observed among rice farmers who had formal education compared to those without formal education. A study in Iraq revealed a higher prevalence of fungal infections in families which had parents with no formal education or low level of education [28], though the study was not amongst rice farmers. Poor personal hygiene and illiteracy are other major factors that influence cutaneous fungal infections in this part of the country. Enlightenment programmes and health education are important in the control of the infection.

The infection was reported more among families living in crowded accommodation and those that interacted with pets/ domestic animals such as dogs, cats, goats, rabbits and poultry birds. The reports of [29] support the findings of this study. It was observed that families share their residential houses with domestic animals such as cats, dogs, goats and poultry birds. Although, rice farmers with no interaction with domestic animals were more infected. Unhygienic conditions among these farmers may be one of the contributing epidemiological factors influencing transmission of infection [30]. Domestic animals which are usually carriers or infected should be bathed with disinfectant at least once a week to reduce the spread of spores. Inadequate and poor infrastructures were also observed in the study areas and could be deduced as one of the factors influencing the prevalence of the infection among the rice farmers.

The results of the study showed that scalp infections were more frequent than nail infections. It was observed that some of the farmers used footwears in the fields whereas most of the farmers did not use hand gloves during their farm work, leading to much higher prevalence of scalp lesions in the study population. This disagrees with the findings of [22] who reported a high rate of infection of the finger and toe nail among rice farmers in Anambra State, Nigeria. This disagrees also with the findings of [12] who reported a high rate of nail infection among paddy field workers in India. Scalp infection was mostly seen in children. This can be attributed to the fact that children are also involved in rice farming.

## CONCLUSION AND RECOMMENDATIONS

In this study, cutaneous aspergillosis and candidiasis caused by *Aspergillus* species and *Candida albicans* were observed to be less prevalent among rice farmers than we had expected. Cutaneous aspergillosis caused by *A. flavus* was observed to be more prevalent and leading to these rice farmers' worse quality of life and economic burden. The confirmation of species of cutaneous *Aspergillus* and *Candida albicans* from the sampled population suggests a relationship that requires further investigation and whose complete understanding has a strong implication for the efficient screening, management, reduction and treatment of the infection among rice farmers, especially in countries like Nigeria where the infection have become a public health problem and have remained endemic. Age, gender, educational status and family size of the farmers and their interactions with domestic animals/pets influenced transmission of the infection.

Although, the present study is a random study that focuses primarily on the prevalence of cutaneous aspergillosis and candidiasis among rice farmers in Ebonyi State, more systematic study covering larger population and over a longer period of time in different geographical locations would give a better insight into the epidemiology of the infection among rice farmers in the country. It is expected that the study will be an eye-opener to the government, non-governmental organizations as well as community-based organizations to execute various intervention programmes like public health education, improved working conditions, housing and sanitary

standards, accessible potable water, protective equipment such as long rubber boots, masks and gloves, periodic surveillance and economic stability to help reduce the burden of the infection among rice farmers in the area.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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