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Original Research Article

Mycological Survey of Unused Tissue Papers in Public Toilets within a University Campus in Port Harcourt, Rivers State, Nigeria

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Abstract

Isolation of fungal species from unused toilet papers is essential, to decipher the public health risk associated with the exposure of these sanitary papers in public toilets, prior to use by students. Unused tissue papers kept at public toilets were sampled from eleven (11) different locations (labelled F1-F11) to cover the various faculties in the University. The fungal counts and characterisation was done following standard microbiological procedures. The result revealed that Total Fungal Count ranged between 3.5±0.4 x 10^4 cfu/g, (obtained from the Faculty of Humanity toilet rooms) and 1.3±0.2 x 10^4 cfu/g, obtained from Faculty of Engineering toilet rooms. Phenotypic characterisation of the fungal isolates revealed the isolates obtained from the various samples included; *Aspergillus niger*, *Aspergillus fumigatus*, *Aspergillus flavus*, Candida species and *Penicillium* species. The percentage occurrence of the isolates showed *Aspergillus niger* as the dominant species (38%), while *Candida* species was the least occurring (17.2%). The study has shown that exposure of unused tissue papers, prior to use in toilets may pose severe health risk to the students. Tissue papers should therefore be kept in sterile packs prior to use in public toilets.

Keywords: Tissue paper, mycological survey, public toilets, University Campus.

1.0 INTRODUCTION

The word 'toilet' is preferred to 'lavatory' because it is understood internationally and encompasses all sorts of facilities ('Ladies' and 'Gents', cubicles, urinals and automatic public conveniences (APCs) [1]. Portable toilets are often provided at festivals and at temporary events for public use [2]. Public toilets can also be seen as comprising both traditional on-street public toilets (usually run by the local authority) and off-street toilets to which the public has access (run by private-sector providers) such as those in shopping malls, sports centres, and railway stations [1].

Toilet rolls are general-purpose, soft paper products for toilet needs of the entire family, most especially for maintaining personal hygiene after human defecation or urination. They can also be used for other purposes, such as food packaging, kitchen cleaning of plates and dishes, cups, cutleries; as sanitary towels / pant liners, and also for emergencies like first aids, to stop / clean light bleedings due to cuts, wounds and bruises.

Toilets harbour microorganisms because of the different microbiomes which are continually being introduced by different users, coupled with resident population growing therein on wet surfaces and organic substances [3]. Studies have shown that fungi are introduced either by means of passive ventilation or by means of ventilation systems [4]. Also, possible sources of biological contamination of indoor air include: people, organic dust, various materials stored in the buildings, and the air inflowing from the ventilation and air conditioning systems [3]. Fungi could be introduced into the toilet by an infected individual's faeces and subsequent infections could be aided by improper hygiene after using the toilet. An investigation carried out by [3], revealed eight fungi species that usually colonize the toilet. They include; *Aspergillus*, *Trichosporonoides*, *Gibellula*, *Beauvaria*, *Stachybotrys*, *Illosporium*, *Botrytis* and *Rhizoctonia*.

There is however scantiness of information regarding the mycological risk associated with tissue paper exposure in public toilets. This present study therefore seeks to present important data set for

research relating to toilet associated infections. The study was therefore aimed at evaluating the mycological risk associated with exposed unused tissue papers in public toilets.

2.0 MATERIALS AND METHODS

2.1 Description of Study Area

The study involved eleven public toilet facilities in the University. The study covered toilet rooms in eleven (11) faculties within the University environment.

2.2 Method of Sample Collection

Unused tissue papers were collected from the different faculties using sterile polytene bags and labelled appropriately. Collected tissue samples were stored at room temperature in a sterile and dry environment.

2.3 Fungal analysis of the tissue paper samples

The Total Fungal (TF) population of the tissue paper samples (F1-F11) were enumerated by transferring 1g of the tissue sample into 9ml each of sterile normal saline, and then serially diluting the solutions via ten-fold serial dilution, to 10^{-3}. Then, 0.1ml of the sample (F1-F11) dilutions (10^{-2} and10^{-3}) respectively, were plated on Sabouraud Dextrose agar (SDA) plates supplemented with chloramphenicol aseptically. Using the spread plate technique, the various samples were plated via aseptic techniques. The inoculated plates were incubated at 37°C for 48 hours. After incubation, colonies were counted and used to calculate fungal population.

Pure fungal cultures were prepared by sub culturing colonial growths onto a freshly prepared SDA plate supplemented with chloramphenicol and incubated at 37°C for 48 hours. Stock cultures were prepared from the pure cultures.

2.4 Identification of Fungal Isolates

2.4.1 Macroscopic Identification

Fungal isolates were identified based on their shape, size, colour, elevation margin, transparency, texture and surface.

2.4.2 Microscopic Identification

Using the wet mount technique for microscopic examination, a small portion of the respective isolates was picked with the aid of a sterile wire loop and placed on a clean grease free glass slide. A drop of lactophenol cotton blue was added and the smear was covered with the aid of a cover slip and examined under the microscope using x10 and x40 objective lenses.

3.0 RESULTS

3.1 Total fungal population of the test samples

Estimation of the total fungal (TF) of the tissue samples labelled F1-F11 is as shown in Table 1. The result showed that highest fungal count was 3.5±0.4 X 10^4 cfu/g, obtained from the Faculty of Humanity toilet rooms. The least count of 1.3±0.2 X 10^4 cfu/g was however, obtained from Faculty of Engineering.

3.2 Frequency of Occurrence and species diversity in the various toilets

Table 2 shows the frequency of occurrence of the isolated fungi from the various samples and their points of collection. It follows that *Aspergillus niger* (38%) was the predominant species, occurring in all the samples.

Based on species diversity as shown in Table3, Faculty of Science had more fungal species, including *Aspergillus niger*, *Aspergillus fumigatus*, *Candida* spp, Penicillium spp. The least number of species was however observed in Faculty of Engineering having only *Aspergillus niger*.

Table-1: Fungal population of the Tissue samples (F1-F11).

Samples Code	Location	THFC X 10^4 cfu/g
F1	Faculty of Management Science	2.9±0.7
F2	Faculty of Engineering	1.3±0.2
F3	Faculty of Agriculture	3.3±0.11
F4	Biology Laboratory	2.6±1.1
F5	ICT	2.0±0.3
F6	Faculty of Education	3.1±0.5
F7	Faculty of Environmental Science	1.5±0.3
F8	Faculty of Science	2.5±0.5
F9	Faculty of Humanities	3.5±0.4
F10	Faculty of Law	2.0±0.2
F11	Shopping Complex	1.5±1.3

TF - Total Fungal Count. Faculty of Management Science (F1), Faculty of Engineering (F2), Faculty of Agricultural Science (F3), Biology Laboratory (F4), ICT (F5), Faculty of Education (F6), Faculty of Environmental Science (F7), Faculty of Science (F8), Faculty of Humanity (F9), Faculty of Law (F10), Shopping complex (F11).

CFU - Colony Forming Unit per Millilitre.
THFC - Total Heterotrophic Fungal Count

Table-2: Frequency of occurrence

Isolate	No	% Occurrence
Aspergillus niger	11	38
Aspergillus fumigatus	5	17.2
Aspergillus flavus	4	13.8
Candida spp	5	17.2
Penicillium spp.	4	13.8
Total	**29**	**100**

Table-3: Distribution of the Fungal Isolates

Sample Code	Location	Fungal Species	No. of Specie
F1	Faculty of Management Science	*Aspergillus niger, Candida* specie , *Aspergillus fumigatus*	3
F2	Faculty of Engineering	*Aspergillus niger*	1
F3	Faculty of Agriculture	*Aspergillus niger, Candida* spp	2
F4	Biology Laboratory	*Aspergillus niger, Candida* spp. Penicillium	3
F5	ITC	*Aspergillus niger, Candida*, spp Penicillium	3
F6	Faculty of Education	*Aspergillus niger, Penicillium*	2
F7	Faculty of Environmental Science	*Aspergillus niger, Candida* spp	2
F8	Faculty of Science	*Aspergillus niger, Aspergillus fumigatus, Candida* spp, Penicillium spp.	4
F9	Faculty of Humanities	*Aspergillus niger, Aspergillus fumigatus, Penicillium*	3
F10	Faculty of Law	*Aspergillus niger, Candida*	2
F11	Shopping complex	*Aspergillus niger, Candida*	2

4. 0. DISCUSSION

The investigation carried out on the various tissue samples ranging from sample F1-F11 showed the presence of fungal organisms. The fungal counts observed in this study may be as a result of the poor state of the toilet facilities in which the samples were collected, which encouraged the growth of fungi. It could also be as a result of the number of individuals that regularly visit the toilet. A previous researcher [5] had opined that poor inter personal hygiene can cause a cross contamination of the toilet paper as seen in the increased fungal count observed in sample F9. Another study by Balanda *et al.*, 2013 [6] observed that the moisture content in the toilet can be a suitable medium for fungal proliferation.

Prominent fungal isolates obtained from the various samples include; Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus, *Candida* spp and *Penicillium* spp. Most of the isolates in this study are in conformity with the isolates obtained by Balanda *et al.* 2013 [6] and Bundesinsitut, 2001 [7].

Gillidon *et al* [5], in their study stated that after proper hand-washing, most people usually dry their hands, especially with toilet papers but there can be transmission of organisms from contaminated toilet rolls. According to Haug *et al*. 2002 [8], fungi, which can be transmitted to humans during usage, have been known to be present on paper products, such as unused paper towels and toilet rolls. It was even reported that average toilet paper dispenser was found to have more organism than the average toilet seat [9].

Aspergillus species were more predominant in the frequency of occurrence than the other isolates. This is in accordance with the experimental run carried out by Balanda *et al.* 2013 [6]. The presence of some pathogens such as *Aspergillu*s sp. and *Penicillium* sp. may raise possible cause for concern as some species of these fungi have been implicated in the production of mycotoxins in food and food products [10]. Most mycotoxins are relatively heat-stable within the conventional food processing temperature range (80–121°C) [10, 11], therefore, little or no destruction occurs under normal cooking conditions, such as boiling. Members of the *Aspergillus niger aggregate* have been implicated in human and animal infections including superficial and local infections (cutaneous infections, otomycosis, tracheobronchitis) [12], infections associated with damaged tissue (aspergilloma, osteomyelitis), pulmonary infections and clinical allergies (allergic bronchopulmonary aspergillosis, rhinitis, Farmers's lung) [10].

Yeast, including *Candida* spp. on the other hand equates to severe vaginal infections leading to yellowish-white discharge with severe itching seen in females of child bearing age [13].

This study does not only implicate tissue paper in the risk of toilet associated infections but also makes the need for proper sanitary practices very critical. Disinfectants should be used in cleaning the toilets rather than regular soaps, in order to kill most of the pathogens [14].

5.0 CONCLUSION

The study has shown that keeping of unused tissue paper in public toilet exposes them to the risk of fungal colonisation thereby posing a public health threat.

The study further showed that *Aspergillus* species predominate in the toilet environment. The presence of candida species further creates more public health concern. It is therefore recommended that tissue papers should therefore be kept in sterile packs prior to use to avoid microbial contamination of the paper product.

REFERENCES

1. Greed, C. (2004, June). Public toilets: the need for compulsory provision. In *Proceedings of the Institution of Civil Engineers-Municipal Engineer* (Vol. 157, No. 2, pp. 77-85). Thomas Telford Ltd.
2. Shamim, M. (2011). The problems of Provision of Public Convenience in Urban Centers. *International Journal of Public Health*, 2(1); 39-41.
3. Ezekanyi, D.N., Nnamani, V.C., Inyang, U.S. (2016). Assessment of Airborne Fungi in an Indoor Laboratory Toilet. *Journal of Pharmacy and Biological Science*, *11*(3); 62-67.
4. Yassin, N.F., Almouqutrea, S. (2010). Assessment of Airborne Bacteria and Fungi in an Indoor and Outdoor Environment. *International Journal of Environmental Science Technology*, *7*(3); 535-544.
5. Gillidon, M.L., Tampid, C.J., Ayers, T. (2005). Fungal Transfer and Cross-Contamination Potential Associated with Toilet Paper Dispensing. *American Journal of Infection Control*, 31:481-492.
6. Balanda, T.F., Maat, L.S., Zinckabja, M.F. (2013). Toilet Microbes Associated with Moisture and Poor Hygiene. *Journal of Microbiology*, *2*(3); 24-40.
7. Bundesinsitut, F.R. (2001). Criteria for Evaluating Sanitary Papers: Standard Specification for Toilet Paper. *Journal of Environmental Science*, (51); 20-28.
8. Haug, T., Soitrand, P., Hanged, S. (2002). Exposure to Culturable Microorganism in Paper Mill and Presence of Symptoms Associated with Infections. American. *Journal of Industrial Medicine*, (41); 498-505.
9. Shanthi, J., Krubakaran, C. T. B., & Balagurunathan, R. (2012). Characterization and isolation of paper mill effluent degrading microorganisms. *J Chem Pharm Res*, *4*, 4436-4439.
10. Okogbenin, O. B., Okogbenin, E. A., Okunwaye, T., Odigie, E. E., & Ojieabu, A. (2014). Isolation of food pathogens from freshly milled palm oil and the effect of sterilization on oil quality parameters. *Journal of Food Security*, *2*(2), 65-71.
11. Gordon, R. E., & Smith, N. R. (1949). Aerobic sporeforming bacteria capable of growth at high temperatures. *Journal of bacteriology*, *58*(3), 327-341.
12. Ohimain, E. I., Izah, S. C., & Jenakumo, N. (2013). Physicochemical and microbial screening of palm oil mill effluents for amylase production. *Greener Journal of Biological Sciences*, *3*(8), 307-318.
13. Sampson, T., & George, A. P. (2019). Evaluating the Risk of Candida albicans Associated with Gestation amongst Women in Port Harcourt, River State, Nigeria. *International Journal of Pathogen Research*, 1-7.
14. Sampson, T., Esheyigba, A. P., & Baridam, S. S. (2019). Bacteriological Assessment of Toilet Seats in a Nigerian University. *Journal of Advances in Microbiology*, 1-11.