Microbiology Research International
Vol. 7(3), pp. 17-23, August 2019
DOI: 10.30918/MRI.73.19.023
ISSN: 2354-2128
Full Length Research Paper

Isolation of microorganisms from infected onions (*Allium cepa*) popularly consumed by low income earners in Ibadan, Nigeria

Salami, O. O.[1]*, Ayanda, O. E.[1], Aluko, O. I.[1], Abimbola, O. T.[2] and Obero, J. O.[1]

[1]Science Laboratory Technology Department (Microbiology Unit), Federal College of Animal Health and Production Technology, P.M.B. 5029, Moor Plantation, Apata, Ibadan, Oyo State, Nigeria.
[2]Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.

Accepted 25 July, 2019

ABSTRACT

Samples of three (3) different varieties of infected onion bulbs were collected from different locations at Oja-oba, Muslim, Oke-ado and Bode markets of Ibadan. Isolation of microorganisms from infected onions (*Allium cepa*) was carried out. The results revealed that the infected onion bulbs were associated with *Escherichia coli*, *Klebsiella* sp., *Salmonella* sp., *Proteus* sp., *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus stolonifer* and *Aspergillus fumigatus*. It was also observed that the market where samples were collected and the variety of onions have no significant effect on the level of bacterial infestation. The mean total bacteria count ranges from 2.2×10^3 to 8.5×10^3. Furthermore, the market where samples were obtained and types of onions had no significant effect on the fungal infestation level. The level of infestation ranges from 1.1×10^1 to 3.1×10^1. *E. coli* had the lowest bacterial prevalence of 8.3% in the study area when compared to *Klebsiella* sp. (33.3%), *Proteus* sp. (30.6%) and *Salmonella* sp. (27.8%). *Aspergillus niger* had the highest fungal prevalence of 44.8% while *Aspergillus flavus*, *Aspergillus fumigatus* and *Rhizopus stolonifer* had prevalence of 26.9, 14.9 and 13.4% respectively. It shows that the different varieties of onions purchased at the selected markets in Ibadan, Oyo State contained different pathogenic microorganisms. Adequate care should be taken in processing onions (*Allium cepa*) to prevent food poisoning.

Keywords: Markets, low income earners, infected onions, microorganism.

*Corresponding author. E-mail: salamiolufunmi@yahoo.com. Tel: +2348028339370.

INTRODUCTION

Onion (*Allium cepa*) is an important vegetable crop commonly grown in the world (Jay, 2005) for consumption and economic value to farmers. It belongs to the family *Alliacea* (Barth *et al.,* 2009) and the crop is grown for its bulbs which are used daily in every home for seasoning and flavouring.

Onion is a valuable ingredient in the diet due to its content of sugars, vitamins and minerals (Ole et al., 2004). There are three (3) varieties of Onions: Red, White and Oriental. Most onion cultivars have been reported to have about 89% water, 4% sugar, 1% protein, 2% fibre and 0.1% fat (Denton and Ojeito, 1990). The consumption of onions in Nigeria has increased tremendously in the recent years probably due to increase in awareness on the health importance (Asagbara and Oyewole, 2002). It can be sautéed to dark brown and served to provide sweet and sour flavour to other foods (Ole et al., 2004).

Red onions sometimes called purple onions are cultivars of the onion with purplish red skin and white fleshy tinged with red. These onions tend to be medium to large in size and have a mild to sweet flavor. They are often consumed raw, grilled or lightly cooked with other foods, or added or as colour to salads. They tend to lose redness when cooked. Red onions are available throughout the year (Hung et al., 2004).

The red colour comes from Anthocyanidins and can be stored 3 to 4 months at room temperature. Red onions are available year round in summer and early fall, when they haven't been in storage for long; they taste sweeter,

with sharpness intensifying through the winter months (Ole et al., 2004).

Oriental onion is an edible species of onion, native to China and cultivated in many other countries. Owing to its very mild and fresh taste oriental onion is often pickled and served as a side dish in Japan and Vietnam to balance the stronger flavour of some other component in a meal. It is used as a folk medicine in tonics to help the intestines and as a stomachic (Ole et al., 2004).

White onion is a type of dry onion that has a pure white skin and a sweet, mild white flesh. This onion is used in Mexican foods or complementing the flavours of other ingredients. Onions (*Allium cepa*) is used widely in seasoning, flavouring and sometimes cured and stored as spicing agent (Frazier and Westhoff, 2005). When improperly stored deteriorates and spoil during preservation which makes it unavailable for use all year round due to the effect of insufficient dryness of bulbs before storage or as a result of little quantity of storage microorganisms present on bulbs going into storage (Banwart, 2001; Charles, 2012).

Pathogens such as: *Pseudomonas* sp., *Staphylococcus* sp. and *Erwinia* sp., were identified with the crop (Dogondaji et al., 2005) and this often reduces germination, and yield of the crop. Pathogen attacks result in a decreased rate of plant photosynthesis (Kocal et al., 2008) and as a consequence yield loss (Berger et al., 2007). Pathogen infection often leads to plant death, the development of chlorotic and necrotic (Kim et al., 2010) lesions and to a decrease in photosynthetic assimilate production. Hence this study was carried out to evaluate the microbiological status of defected onions consumed by low income earners (Frazier and Westhoff, 2005; Banwart, 2001). The aim of this work is to isolate, identify and characterize the different micro-organisms that are associated with spoilage of onions and to categorize the pathogenic micro-organisms associated with the spoilage of onions.

Aim and objectives

The aim of this study is to isolate microorganisms from infected onions (*Allium cepa*) popularly consumed by low income earners in Ibadan, Nigeria with the under listed objectives:

i. To isolate the microorganisms causing spoilage in the selected onions
ii. To identify the microorganisms isolated in (i) above
iii. To characterize the microorganisms identified in (ii) above using biochemical test

Justification

It is an investigation to checkmate the public health significance on people by determining the risk posed to the consumers of spoilt onions and to suggest control measures and possibly solutions in avoiding the spoilage and its storage.

MATERIALS AND METHODS

Sample collection

Three different varieties of infected onions (Red, Oriental, and White) were randomly sampled from four different markets (Bode, Oja-oba, Oke-Ado and Muslim) all in Oyo State. The samples were kept in sterile labeled polythene bags and taken to the Laboratory for further analysis (Appendix).

Culturing and isolation of organisms

An appropriate section of 1.0 g were cut from each of the sample purchased from the markets with a sterile scalpel knife and was homogenized. The resultant homogenate was added to 10 ml of distilled water in a test tube and diluted serially. This was done to thin out the microbial load in the samples. Sterile pipettes were used to take 1ml of the serially diluted samples and plated on different media using spread plate method.

All culture media were prepared according to manufacturer's instruction. MacConkey Agar (Mcc) and Nutrient Agar (NA); 25 g of the agar was weighed and dispensed into a sterile conical flask, 150 ml of distilled water was also measured and added to the agar. The mixture was homogenized and heated in a microwave for 10 min, then sterilized at 121°C for 15 min and allowed to cool before pouring. It was incubated at 35°C for 24 h for bacterial count and observes the morphological features of bacterial colony while Saboraud Dextrose Agar (SDA) for fungal growth was incubated at room temperature (25°C) for 72 h. Sub culturing of different distinct colonies from bacteria growth was done on Nutrient agar and stored at 4°C for biochemical test.

All the bacterial isolates were identified on the basis of colony characteristics, morphological and biochemical reactions as described in Bergeys' Manual of Determinative Bacteriology (Buchanan and Gibbons, 1974).

Fungi isolates were observed under the microscope using the wet mount method. A loop of the isolated colony was teased out onto a grease free slide and a drop of lacto phenol cotton blue was added and covered with cover glass and the fungus was examined microscopically. It was viewed under the microscope at ×40 objective lens. Colonies of fungi were observed on plate for colour, appearance and under the microscope for its spore type and nucleus which were compared with the diagram and structure of known fungi using identification keys of Ellis et al. (2007) and Campbell et al. (2013).

RESULTS AND DISCUSSION

From table 1, the mean total bacteria count for red onion, obtained at Bode, Oja-Oba, Oke-Ado and Muslim markets were not significantly different from each other ($p \leq 0.05$). Also, the mean total bacteria count obtained on oriental onions at Bode, Oja-Oba, Oke-Ado and Muslim markets were not significantly different from each other ($p \leq 0.05$). The mean total bacteria count obtained on white onions at Bode, Oja-Oba, Oke-Ado and Muslim markets

Table 1. Mean total bacterial count (cfu/g) isolated from different varieties of onion (*Allium cepa*) in different markets in Ibadan.

Market	Red ×10^3	Oriental ×10^3	White ×10^3
Bode	3.9 ± 0.5^{a*}	3.2 ± 0.1^{a*}	2.2 ± 0.2^{a*}
Oja-Oba	8.5 ± 0.1^{b*}	5.3 ± 0.3^{b*}	4.2 ± 0.2^{b*}
Oke-Ado	6.6 ± 0.2^{b*}	5.8 ± 0.2^{b*}	4.5 ± 0.3^{b*}
Muslim	4.3 ± 0.3^{b*}	6.1 ± 0.3^{b*}	4.2 ± 0.2^{b*}

* indicates no significant difference down the column
Same lowercase superscripts indicate no significant difference across the row.

Table 2. Mean total fungal count (cfu/g) isolated from different varieties of onion (*Allium cepa*) in different markets of Ibadan.

Market	Red (×10)	Oriental (×10)	White (×10)
Bode	2.5 ± 0.5^{aB}	2.4 ± 0.5^{aB}	1.8 ± 0.3^{aB}
Oja-Oba	1.2 ± 0.2^{aB}	2.5 ± 0.1^{aB}	2.5 ± 0.2^{aB}
Oke-Ado	1.1 ± 0.3^{aB}	2.2 ± 0.3^{aB}	1.9 ± 0.1^{aB}
Muslim	2.2 ± 0.1^{aB}	3.1 ± 0.1^{aB}	2.4 ± 0.1^{aB}

Same lowercase superscripts indicate no significant difference across the row
Same uppercase superscripts indicate no significant difference down the column.

were also not significantly different from each other ($p \leq 0.05$). These indicate that the market where onion samples were obtained has no significant effect on the level of bacterial infestation of onions.

The mean total bacteria count obtained from red onion, oriental onions and white onion at Bode, Oja Oba were not significantly different from each other ($p \leq 0.05$). The mean total bacteria count for red onion, oriental onions and white onion obtained at Bode, Oke-Ado and Muslim markets respectively were not also significantly different from each other at each of the market ($p \leq 0.05$). This implies that the variety of onions also has no effect on the level of bacterial infestation. From this Table 1, it could be observed that, neither the market nor species of onions has effect on the bacterial infestation level. The level of infestation ranges from 2.2×10^3 on white onion obtained at Bode market to 8.5×10^3 on red onion obtained at Oja-Oba market.

From Table 2, the mean total fungal count obtained reveals that, there were no significant differences ($p \leq 0.05$) in the values obtained from different type of onions. Also, there was no significant difference ($p \leq 0.05$) in the mean total fungal count at different market where samples were obtained. This implies that neither the market nor species of onions has effect on the fungal infestation level. The level of infestation ranges from 1.1×10^1 on red onion obtained at Oke-Ado market to 3.1×10^1 on Oriental onion obtained at Muslim market.

This agrees with the reports of other researchers (Muhammad et al., 2004; Dimka and Onuegbu, 2010) that fungi constitute a menace in the storage of many agricultural commodities including fruits, vegetables and nuts.

From Table 3, four bacteria isolates were identified; they are *Klebsiella* sp., *Proteus* sp., *Salmonella* sp., and *Escherichia coli*. *Klebsiella* sp. has the highest prevalence of 33.2% in the study area. The prevalence of *Proteus* sp. was 30.5%, *Salmonella* 27.9% while *E. coli* had the least prevalence of 8.3% in the study areas.

The prevalence of *Klebsiella* sp. was 8.3% at each market where samples were taken. *Proteus* sp. has prevalence of 5.6% at Bode market while having 8.3% in the other three markets respectively. *Salmonella* sp. has prevalence of 11.1% at Oke-Ado market while having 5.6% in the other three markets respectively. *E. coli* was isolated only at Muslim market; it was not isolated from Bode, Oke-Ado and Oja-Oba markets (Figures 1 and 3).

From Table 4, four fungi strain were isolated; *Aspergillus flavus*, *Aspergillus fumigatus, Rhizopus stolonifer and Aspergillus niger. A. niger* had the highest prevalence of 44.8% while *Aspergillus flavus*, *A. fumigatus* and *R. stolonifer* had prevalence of 26.9, 14.9 and 13.4%, respectively. Prevalence of each of the four isolated fungi species were lowest at Muslim market with *Aspergillus niger* having 7.5%, *Aspergillus flavus* 6.0%, while *A. fumigatus, R. stolonifer* had 1.5%, respectively (Figures 2 and 4).

Prevalence of each of the four isolated fungi species were highest at Bode market with *A. niger* having 17.9% , *A. flavus* 7.5%, *A. fumigatus,* 6.0% and *R. stolonifer* having 4.5%. This agreed with the findings of Adebayo and Diyaolu (2003).

R. stolonifer (4.47%) had the highest number of isolate from Bode market and Oke-Ado markets respectively as

Table 3. Prevalence of bacteria isolates from different onion varieties (*Allium cepa*) in different markets in Ibadan.

Isolates	Total no. of isolates	Prevalence of isolates (%)	Prevalence of isolates at each markets (%)			
			Bode	Oke-Ado	Oja-Oba	Muslim
Escherichia coli	3	8.3	0	0	0	8.3
Salmonella sp.	10	27.9	5.6	11.1	5.6	5.6
Proteus sp.	11	30.5	5.6	8.3	8.3	8.3
Klebsiella sp.	12	33.2	8.3	8.3	8.3	8.3

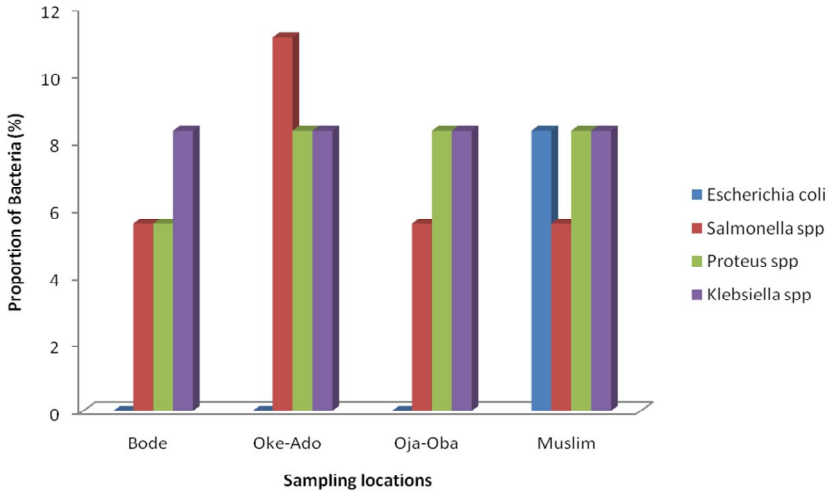

Figure 1. Prevalence of bacterial isolates.

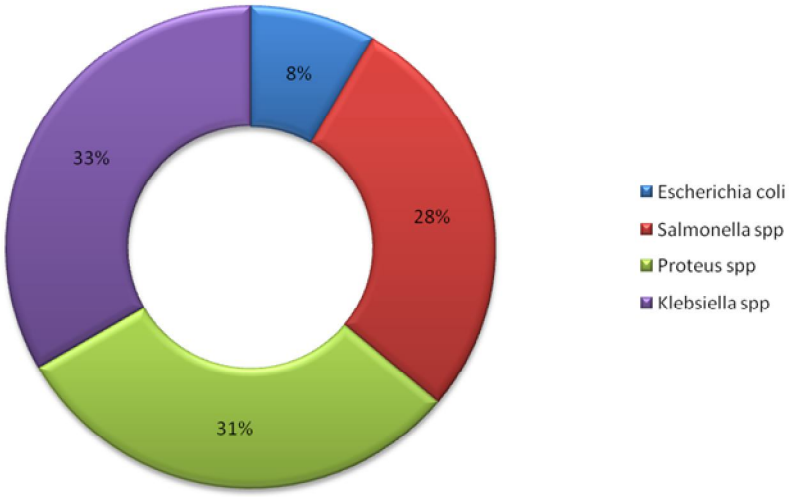

Figure 3. Pictorial prevalence of bacterial isolates.

reported by Dimka and Onuegbu, 2010.

From this result, it shows that onions purchased from Bode, Oke-Ado, Oja-Oba and Muslim markets in Ibadan were contaminated with bacterial and fungal isolates which were *Klebsiella* sp., *Proteus* sp., *Salmonella* sp., *E. coli*, *A. flavus*, *A. fumigatus, R. stolonifer* and *A. niger.* Different species of onions have effect on the level of both bacterial and fungal infestation. Also, location where samples were obtained did not also have effect on the degree of infestation of bacteria and fungi. However, Bode market has the highest prevalence of fungal infestation while Muslim market has the least. Therefore,

Table 4. Prevalence of fungi Isolates from Onion (*Allium cepa*) in different markets in Ibadan.

Isolates	Total no. of isolates	Prevalence of isolates (%)	Prevalence of isolates at each markets (%)			
			Bode	Oke-Ado	Oja-Oba	Muslim
Aspergillus niger	30	44.8	17.9	9.0	10.4	7.5
Aspergillus flavus	18	27.0	7.5	6.0	7.5	6.0
Aspergillus funmigatus	10	15.0	6.0	4.5	3.0	1.5
Rhizopus stolonifer	9	13.5	4.5	4.5	3.0	1.5

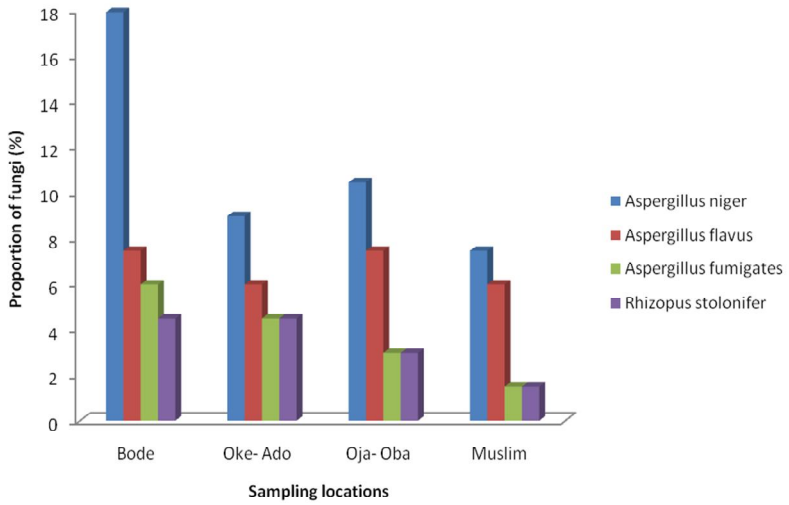

Figure 2. Prevalence of fungal isolates.

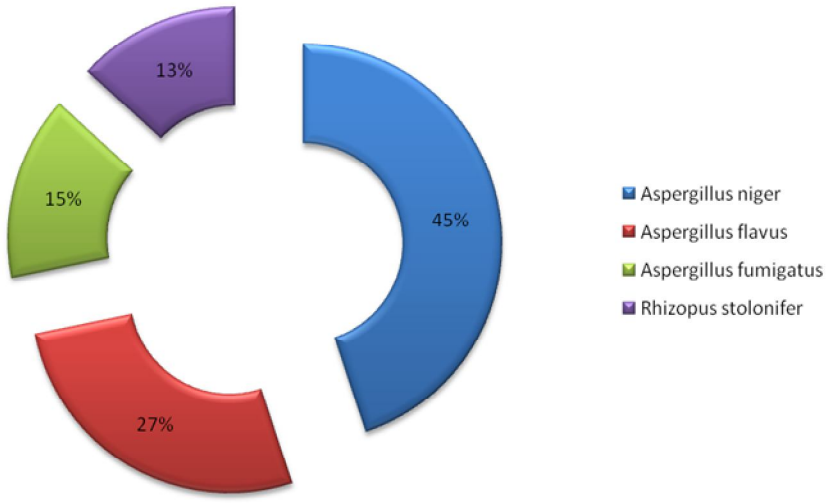

Figure 4. Pictorial prevalence of fungal isolates.

table 5 described the fungi seen and isolated in the selected markets.

E. coli had low prevalence of 8.3% in the studied area when compared to *Klebsiella* sp. (33.3%), *Proteus* sp. (30.6%) and *Salmonella* sp. (27.8%).

This contamination could be as a result of poor packaging, transporting, and storage. Onions packed in nylon should immediately be spread out in air after

Table 5. Morphology description of isolated fungi from onion (*Allium cepa*) in different markets in Ibadan.

Organisms	Colour	Appearance	Spore type	Nucleus
Aspergillus niger	Surface is black while the underneath is white to yellow	Filamentous with branching mycelia	Long, smooth, brown or yellow conidiospores	Haploid, multinucleated
Rhizopus stolonifer	Dark or black with patches	Filamentous with cottony mycelia	Sporangiospore	Multinucleated
Aspergillus flavus	Yellow-green while the underneath is gold to red-brown	Filamentous with branching mycelia	Colourless rough conidiospores	Multinucleated
Aspergillus fumigatus	Blue-green on its surface and white underneath	Filamentous with branching mycelia	Short, smooth grey-green conidiospores	Haploid

purchase to prevent spoilage caused by the heat generated from the polythene bags. Spoilt onions sold in the market should be avoided. However, adequate care should be taken when processing onions to prevent food poisoning. Further studies should be carried out on onion handling and quality control practices in the markets with a view to avert post-harvest losses and food poisoning.

CONCLUSION

From the results of this study, it can be concluded that all the onions purchased from different markets had high degrees of microbial isolates; conclusively, most of the samples were contaminated with micro-organisms; which is not quite suitable for the consumption of people. It also shows that onion bulbs could be a reservoir for microorganisms such as fungi and bacterial and it could also be as a result of exposing the onions to poor environment like poor packaging (which may increase its temperature and humidity) transporting, and storage.

RECOMMENDATION

It is therefore recommended that onions packed polythene bag should immediately be spread out in air for proper and good ventilation after purchase to prevent spoilage caused by the heat generated from the polythene bags.
Good storage facility should be made available
Spoilt onions sold in the market should be avoided and its consumption discouraged.
However, adequate care should be taken when processing onions to prevent food poisoning. Further studies should be carried out on onion handling and quality control practices in the markets with a view to avert post-harvest losses and food poisoning.

ACKNOWLEDGEMENT

The authors wish to thank Professor Olukayode Asaye of Institute of Agricultural Research and Training, Moor plantation for his contribution and fatherly advice.

REFERENCES

Adebayo LO, **Diyaolu** SA, **2003**. Mycology and spoilage of cashew nuts. Afr J Biotechnol, 2: 369-373.
Asagbara A, **Oyewole** OB, **2002**. Fermentation studies on carrot juice processed to table wine. Nig Food J, 20: 74-77.
Banwart GJ, **2001**. Bacteria as food spoilage organism basic food microbiology. Avi Publishing co. Westport.com. pp: 119-125.
Barth M, Hankinson TR, Zhuang H, Breidt, F, **2009**. Microbiological spoilage of fruits and vegetables. W. H. sperber, M.P. Doyle (eds.) compendium of the microbiology spoilage of foods and beverages, food microbiology and food safety. C springer science + business media, LLC, pp 35-183.
Berger S, Sinha AK, Roitsch T, **2007**. Plant physiology meets phytopathology: plant primary metabolism and plant-pathogen interactions. J Exper Bot, 58: 4019-4026.
Buchanan RE, **Gibbons** ME, **1974**. Bergeys Manual of Determinative Bacteriology. 8th ed. Williams and Wilkins, Baltimore. Pp: 305-318.
Campbell CK., Johnson EM, David W, Warnock DW, **2013**. Identification of pathogenic fungi. 2nd ed. Wiley-Blackwell, Chichester, West Sussex. pp 337.
Charles PD, **2012**. *Escherichia coli* 0157:H2. www.medicinenet.com. Accessed. August 2013.
Denton L, **Ojeifo** IM, **1990**. Onion production practices and their Improvement in Nigeria. Onion Newsletter for the Tropics, pp: 11 – 13.
Dimka SON, **Onuegbu** BA, **2010**. Mycoflora of copra and effect of brining on some properties of copra in Nigeria. Agric Biol J North Am, 1(3): 391-394.
Dogondaji SD, Baba KM, Muhammad I, Magaji MD, **2005**. Evaluation of onion storage losses and implication for food security in Sokoto Metropolis. Bull Sci Assoc Nig, 26:10-14.
Ellis D, Davis S, Alexiou H, Handke R, Bartley R, **2007**. Descriptions of Medical Fungi. 2nd ed. University of Adelaide, Adelaide, Australia. pp:8-124.
Frazier WC, **Westhoff** DC, **2005**. Food microbiology. Mc Graw-Hill Publishing Company Ltd., New Delhi pp. 217-240.
Hung HC, Joshipura KJ, Jiang RH, Hunter D, Smith SA, **2004**. Fruits and vegetables intake and risk of major chronic diseases. J Nat Cancer Instit, 95(10): 157-164.
Jay MJ, **2005**. Modern Food Microbiology. 4th ed. Chapman and Hall. New York. p. 187.
Kim YM, Bouras N, Kav NN, Strelkov SE, **2010**. Inhibition of photosynthesis and modification of the wheat leaf proteome by Ptr ToxB: a host-specific toxin from the fungal pathogen *Pyrenophora tritici-repentis*. Proteomics, 10: 2911-2926.
Kocal N, Sonnewald U, Sonnewald S, **2008**. Cell wall-bound invertase limits sucrose export and is involved in symptom development and inhibition of photosynthesis during compatible interaction between tomato and *Xanthomonas campestris pv vesicatoria*. Plant Physiol, 148: 1523-1536.

Muhammad S, Shehu K, Amusa NA, **2004**. Survey of the market diseases and aflatoxin contamination of tomato (*Lycopersicon escolentus* Mill) Fruits in Sokoto, Northwestern Nigeria. Nutr Food Sci, 34(2): 72-76.

Ole H, Torben L, Lars PC, Ulla K, Nazmul H, Shakuntala HA, **2004**. Contents of iron, zinc and β ca-rotene in commonly consumed vegetables in Bangladesh. J Food Composit Anal, 17:587-595.

Citation: Salami OO, Ayanda OE, Aluko, OI, Abimbola OT, Obero JO, 2019. Isolation of microorganisms from infected onions (*Allium cepa*) popularly consumed by low income earners in Ibadan, Nigeria. Microbiol Res Int, 7(3): 17-23.

APPENDIX

Red onion

White onion

Oriental onion