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ISOLATION AND CHARACTERIZATION OF MICROORGANISMS IN KITCHEN CUTTING BOARD

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Abstract:
Kitchen cutting board is a durable board in which materials are placed for cutting. Eight (8) samples were collected from different kitchen cutting board in Ozoro metropolis of Delta state to ascertain the level of microbial contamination. The microorganisms isolated from kitchen cutting board are Escherichia coli, Staphylococcus aureus, Klebsiella spp, Candida albican, Aspergillus niger, Penicillium spp and Mucor spp. The total bacteria count ranges from $0.3*10^1$ to $9.2*10^3$ with E.coli having the highest percentage occurrence of 41.7% while Klebsiella spp. has the least percentage occurrence of 25.0% in all samples. The fungi count ranges from $0*10^1$ to $3.1*10^3$ with Candidas albican having the highest percentage occurrence of 45.5% while Mucor spp has the least percentage occurrence of 9.1%. The result obtained from this study shows that kitchen cutting board is contaminated with pathogenic microorganisms. Proper washing of the kitchen cutting board before and after use should be practiced.

Key words: *Isolation, Characterization, Kitchen, Microorganism, cutting board.*

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INTRODUCTION:

A kitchen cutting board is a durable board on which materials are placed for cutting.

The kitchen cutting board is commonly used in cutting vegetables, and some species. Other types exist for cutting raw materials such as leather or plastic. Kitchen cutting boards are often made of wood or plastic and come in various sizes and widths. There are also cutting board made of glass, steel or marbles, which are easier to clean than wooden or plastic ones such as nylon or coria, but tend to damage knives due to their hardness, rough cutting edge-such as serrated knives-abrade and damage a cutting surface more rapidly [1],. Most of what we eat will pass across its surface route to pan or oven. Now a new survey has revealed the chopping board to be one of the dangerous utensils, harbouring 200 more fecal bacterial than the average toilets pepsins[2] the research commissioned by the global hygiene council (GHC) found 40 percent of all food poisoning cases are caused by poor hygiene in the home. Almost half of all frequently touched items in the home including; chopping boards are contaminated with harmful bacterial including *E. coli* [3].

According to GHC 2000, kitchen cutting board hygiene expert and GHC(Global hygiene council) representative said, in all surveys that have been done focusing in the homes, chopping board come out badly, the reason is because don't clean them properly.

Chicken, turkey and fowls are associated with Shigella and Campylobacter. These are bacteria that cause diarrheal, cramping and fever. Most meat could be contaminated with Toxoplasmosis. This is a parasitic disease dangerous to both women and foetuses [4]. Contaminated vegetables and fruits can carry a variety of organism and parasite depending on where they were grown and how they were processed, derived from CDS.

The objectives of this study are to isolate and characterize microorganisms in kitchen cutting board. This study will consider the importance of hygiene in our kitchen.

MATERIALS AND METHOD:

Study Area

This study was conducted in Ozoro Delta State of Nigeria. Ozoro is in Isoko North Local Government Area of Delta state. The people are Isoko speaking and hospitable. Their main activity is food crop farming accompanied by some hunting. They are also engaged in trade of foods crop for cash to meet other basic household needs. The region experience higher rainfall and humidity most of the years.

Study Size

The samples used for this research were eight (8) samples and they were obtained from different residential apartment in the study area

Sample Collection

Sterile swab stick was used to swab the cutting board of different residential apartments. The samples were collected using sterile swab stick and normal saline was added to it properly covered at the place of collection and the samples were labelled A-H. The samples were all transported to the laboratory, where analysis was carried out.

Materials

The materials used in this study includes laboratory coat, gloves swab stick, incubator, autoclave measuring cylinder (50mc, 250mc, 500ml) and beakers (50ml, 200ml, 500ml) petri dishes test tubes conical flask, Bunsen burner, nutrients agar, triple sugar iron agar, citrate agar, peptone water, wire loop, and honey.

Sterilization of Glass Wares

The glass wares that were used for this project were washed with detergent rinsed thoroughly and sterilized using autoclave at 121^0C for 15 minutes

Methods

Isolation of Test Organism

Three agars were used and they include Nutrient agar, Sabouraud Dextrose agar and MacConkey agar. Media prepared was according to the manufacturer instruction and then used for isolation of bacteria and fungi. Pure isolates where identified according to their morphological characteristics and reactions to biochemical test

Characterization and Identification of Fungi

Morphological and microscopic characteristics of the culture were used for the identification of the isolate following standard references. The colony morphology used includes; color of spores, present or absent of pigmentation, elevation, and nature of mycelia. Microscopic Characteristics used for the identification include the type and the shape of asexual and sexual spores, present or absent of cross walls in hyphae, presence or absence of sterigmata and the sporangiosphores

Morphological Characteristics Gram Staining

Smear of each bacterial isolate was made on a grease free clean glass slide with a drop of normal saline air dried and heat fixed by quickly passing the slides over flame. The Smear was flooded with crystal

violet for one minute (1 minute) then washes add Lugol's iodine solution for 1 minute and then flooded with water again. The slide was then flooded with safranine red for one minute to counter stain and washed off with water, dried and examined under the microscope using oil immersion and x100 objectives.

Biochemical Test
The biochemical analyses carried out were in accordance with procedures reported by (Cheesbrough, 2002) [5].

Citrate Test
The bacteria isolate were tested for their ability to utilized citrate as the sole carbon source. Simmons citrate medium was used.
Bacteria isolates were inoculated into Simmons citrate medium in test tubes and incubate at 37^0c for 24-28 hours. The culture media was observed for a color change from green to blue. Positive shown no growth with intense blue color, while negative test showed no growth and the color of the medium remained green (Bello, 2000)

Triple Sugar Iron Agar Test (TSI)
Bacterial isolated were stabilized into TSI slant media and also streaked on the surface slant after while the medium was incubated at optimal temperature of 37^0c for 24 hours. The TSI slant medium was used to check for the present of the following

Gas: if bubble is present in the media (Gas positive)

H_2S: if black is present in the media (H_2S positive)

Lactose: if the top of the media turn from pink to yellow (lactose positive)

Glucose: if the bottom of the media turn from pink to yellow (glucose positive)

Catalase Test
This test detects the presence of catalase enzymes when present in a bacterium, it catalyze the breaking down of hydrogen peroxide with the release of oxygen as bubble.

$$2H_2O_2 \xrightarrow{\text{Catalase}} 2H_2O + O_2$$

With a wire loop, a colony was picked from the pure culture and was transferred to the Centre of a glass slide 1-2 drops of 3% hydrogen peroxide was added to the bacterial isolates immediate production of bubbles indicated positive result and if no bubble indicated negative.

Indole Test
This test demonstrates the ability of certain bacterial to decompose the amino acid tryptophan to indole production.Bacteria isolates were incubated into peptone water medium contained in sterile test tubes then incubated at 37_0c for 48hurs. After the incubation period about 3 drops of Kovac's indole reagent was added to the peptone water culture. The bottles were shaken thoroughly and allow standing and observed for colour development. A red colour ring at the interface of the medium denotes a positive result. And if the isolate is negative, the reagent layer will remain yellow or slightly cloudy.

RESULTS:

Table 1: Shows the identification and characterization of bacteria isolates

Isolated	Gram stain	Morphological characteristic	Citrate	Oxide	Catalase	Indole	glucose	Latose	H2S
E.coli	GNSG	Rods	+	+	+	-	+	+	+
Klesiella spp.	NSGP	Rods	+	-	+	+	+	+	+
Staphylococcus aureus	S	cocci	-	-	-	-	+	+	-

Key
+ represent a positive reaction
-Represent a negative reaction

Table 2: shows the total bacteria count isolated from the sample

Samples	Total bacteria count
A	$0.4*10^1$
B	$0.5*10^1$
C	$1.0*10^2$
D	$0.9*10^1$
E	$4.2*10^3$
F	$9.2*10^3$
G	$0.3*10^1$
H	$0.3*10^2$

Table 3: Shows the percentage occurrence of bacteria isolates

Isolates	% of occurence
E.coli	41.7
Staphylococus aureus	33.3
Klebsiella spp.	25.0
Total	100

Table 4: Cultural and microscopic characteristics of fungi isolates from kitchen cutting board

Cultural characteristics		Microscopic characteristics	Organisms identified
	Black fluffy growth with white edges	Thick *septate hyphae* with conidia borne in chains from the *sterigmata*	*Aspergillus niger*
	White, heavy, wooly, fluffy growth covering entire plate	Thick *non-septate hyphae* with dark *sporangiospores*	*Mucor spp*
	Blue-green fluffy growth on plate	Blue-green *conidiospores* borne in multilink chains	*Penicillium spp*
	The colony are cream without profuse growth.	Hyphae and conidiospores are non-septate	*Candida albicans*

Table 5: shows total heterotrophic count of fungi isolates

Samples	Total fungi count
A	$1.3*10^3$
B	$0*10^1$
C	$2.5*10^3$
D	$2.1*10^3$
E	$2.9*10^3$
F	$3.1*10^3$
G	$0.3*10^1$
H	$1.8*10^3$

Table 6: Shows the percentage of Occurrence of fungi isolates

Fungi isolates	Percentage(%)of occurrence
Candida albicans	45.5
Aspergillus niger	27.2
Penicillium species	18.2
Mucor spp.	9.1
Total	100

DISCUSSION:

Unhygienic kitchen is a complex ecosystem of microorganisms which could be transient or resident (Leifer and Nester, 2008).

The result obtained from this study showed that mutilated kitchen cutting boards are contaminated with microorganisms. This brings to the mind the question how microorganisms are found in kitchen cutting board. They provide favourable conditions such as substrates acquired from the kitchen cutting board and due to handling as well as improper washing before and after usage [2]. This suggests that human are the major source of microorganism in the kitchen cutting board. The kitchen cutting board could have been colonized when not allowed to dry under normal temperature in the kitchen.

Cutting board is mostly contaminated if it is not properly washed after usage. The result from this study shows that E. coli has the highest percentage occurrence while *Klebsiella spp* has the least percentage occurrence (table 3). *E.coli*, *Staphylococcus aureus* and *Klesiella spp.* were the three identified bacterial isolates that colonized the cutting board.

Staphylococcus aureus was isolated from the kitchen cutting board (table1). The result were in agreement with other studied, that kitchen cutting board are highly contaminated with raw food such as fruit, meat, vegetables and others followed by cooked food and juice (Yasin and Tallow, 2012). Exposure to pathogens may occur by their indirect or direct contact with contaminated object or indirect through airborne particles. They also indicate that some bacteria such as *E.coli* and *Staphylococcus spp*. could survive on hard, sponges and other object for up to several days after contact (Hazeleger and Burrow 2002).

Also from the result of this study four fungi species were isolated; *Candida albican*, *Aspergillus niger*, *Penicillium spp.* and *Mucor spp.* (table 4) .Candida albican has the higest percentage occurrence of 45.5% while *Mucor spp*. has the least percentage occurrence of 9.1%. Low hygiene cutting board can cause the transfer of *Candida albican* from kitchen cutting board to human system. This fungus is found in almost everyone, located in their gut.

CONCLUSION:

The result of the study report suggested that kitchen cutting boards might be a possible vehicle of microorganisms. Unhygienic cutting board carries a lot of pathogenic micro organisms (fungi and bacteria). There should be proper handling of food stuff that comes in contact with the cutting board to prevent contamination of food poisoning microorganisms.

Recommendations

Kitchen deserves special attention. The practice of using hand in cleaning of cutting board instead of proper washing enteric pathogens. Strategies to reduce contamination of kitchen cutting boards, especially personal hygiene should be noted. Other recommendation are the washing of kitchen cutting board before and after usage, proper washing of vegetables before slicing with the cutting board should be observed . Finally, it is recommended that personal and domestic hygiene should be practiced.

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