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Bacteriological Analysis of Deteriorated Persea Americana (Pear Fruit)

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Abstract

The Avocado pear (Perseaamericana) is a member of the family Lauraceae. This fruit has a short shelf-life, it can be averagely stored 3-6days before spoilage. This study was aimed at analysing bacteriologically deteriorated Avocado pear (Perseaamericana). The sample was collected and allowed to spoil before serial dilution was conducted. There was growth of five (5) bacteria organism that was identified. Bacillus species had very high growth (5.1 x 10^5cfu/g) followed by shigellaspecies, staphylococcus aureus which were moderate and salmonella sp. which was few. Biochemical analysis was carried out, which include gram staining, catalase test and Indole test. This research work should be encouraged for more parasitic organisms such as fungi and virus contamination. This project work was achieved.

INTRODUCTION

The Avocado pear *(Perseaamericana*) is a member of the family *Lauraceae*. It is a tropical and sub-tropical fruit tree, originated in Central America or adjoining regions of North or South America (Ayala *et al*., 2021). It has now spread too much of the near tropical world. The avocado is limited especially by its climatic requirements, with their race differences. It is also highly susceptible to drought injury. But excess soil moisture is equally fatal, encouraging the dread phytophthoraroot rot in most of the world, and where the fungus is unknown. Despite the high water activityof most fruits, the pH leads to their spoilage being dominated by fungi, both yeast and moulds. Other common diseases of pears include the black spot or scab caused by as comycetes (Barry, 2021).

Avocado fruits has being estimated that one fourth of all produced harvested is not consumed before spoilage(Chen *et al*., 2018), spoilage of fresh fruits usually occurs during storage and transport and while washing to be processed unlike many other foods fruits after picking and before processing the products and the normal ripening process complicate an independent discussion of the microbiological spoilage problems are really market diseases of these products: Shape, varies from slightly obligate to highly elongate, an intermediate oval or pear shape is preferred. Yellow to purple or black (Chikwe, 2020).

This fruit has a short shelf-life, it can be averagely stored 3-ódays before spoilage. The poor shelf life of the fruit has led to its high perishability. The fruit is a prime, characterized by a central core surrounded by edible fleshy layer (Barry, 2021). The avocado fruit has a puppy mesocarp of thickness, 7cm-20cm long, weighs 100g - 1000g and has a large central seed, 5cm -6.4cm long. The skin texture is finely pebbled and dull green when ripe. The oils from the pulps and seeds of avocado fruit can be used in foods, pharmaceutical and cosmetic manufacturing as well as numerous industries uses. They are rich in mono saturated fatty acids and are comparable to other currently used vegetable oils (Lopez *et al*., 2019).

Avocados are a source of vitamins C, E, K, and B6, as well as riboflavin, niacin, folate, pantothenic acid, magnesium, and potassium. It provides a substantial amount of monounsaturated fatty acids and is rich in vitamins and minerals. Loaded with folic acid, avocado pears does not only help in forming the nervous system of the unborn child, it alsoensures healthy brains, cells, and blood. In addition, it helps to reduce the cholesterol levels ofthe pregnant mother and reducing the risk of depression as well. Avocado fruit is a major and cheap source of nutrients containing (2g), moisture (72.23g), fibre (6.7g), fat (14.66g), carbohydrate (8.53g) and 160 kcal per 100g (Solazer*et al*., 2018).

Micro-organisms influences the spoilage, thus bacteria soft rot is widespread for the most part among the fruits is limited to those that are not highly acidic. Because most fruits are deficient in B vitamins, mold is the most common causes of spoilage. The compositions too, must determine the particular kind of molds that mostly grow in them, thus avocado support a large, variety of fungal spoilage organisms and other kinds comparatively few bacteria like *Enterobacter, Citrobacter, Acinetobacter, Klebsiella, Aeromonas and Micrococcus sp. Enterobacter sp., Bacillus sp.* (Ohr*et al*., 2023). This study is aimed at analyzing bacteriologically deteriorated avocado pear (*Perseaamericana*).

MATERIALS AND METHODS

SAMPLE COLLECTION

Two samples each were purchased at Bida markets. The avocado pear samples collected was fresh, undamaged, firm, healthy and ripe. The samples was dispensed into clean bags and then brought to the laboratory. The samples were left free of dust, insect and were under room temperature for between 5-6 days to undergo natural process of spoilage before being used in the study.

MEDIA PREPARATION

The media was prepared strictly according to manufacturer's instruction. They were all sterilized by autoclaving at 121°C for 15 minutes. After sterilization the agar was allow to cool down to a temperature of 40°C into appropriate Petri-dishes. Culture media used were nutrient agar for bacteria and SauboraudDextrose Agar (SDA) for fungi.

SAMPLE ANALYSIS

Samples were blended using a sterile blender. A homogenate of each sample was made by blending ten grams in 25ml of sterile water and then the sample was blended. Serial dilutions ofup to 10^{-1} – 10^{-5} were made in sterile test tubes by several transfers of 1 ml of previously diluted samples from one dilution tube to 9m1 of sterile water in another tube.

INOCULATION AND INCUBATION

After preparation of serial dilution up to io'-iíi, then imi of serially diluted avocado pear sample was pipetted out to each serially marked petri-dish. Nutrient and Sauboraud Dextrose Agar was poured into appropriately marked plates. The nutrient agar plate was then incubated at a temperature of 37°C for 24hours, while the SDA was left at room temperature for 5 days.

ISOLATION OF BACTERIA

Distinct colonies from the SDA and nutrient agar were sub-cultured into freshly prepared agar using aseptic techniques to prevent contamination. The plates were incubated at room temperature for 37°C for 24 hours. The developed colonies was counted and colonies forming units was calculated and recorded. The colonies was purified and then later stored in nutrient agar slant in refrigerator (4°C) for characterization.

CULTURAL CHARACTERISTICS

Colour, margin and shapes of the bacteria on the media was observed and recorded.

GRAM STAIN

The gram stain was carried out on 24 hours cultures. A smear of each of the bacterial isolates was made on clean grease free slide and heat- fixed using flame. Crystal violet stains (0.3%w/v) was and allowed to stand for one minute. The stain was washed off with distilled water. Iodine (0.4% w/v), a mordant was added and allowed to stand for one minute before being rinsed off with distilled water. Ethanol (95% w/v), a decolourizer was then added and allowed to stand for 30 seconds before being rinsed off with distilled water and then counterstained with thesecondary stain, safranin (0.4%w/v) and allowed to stand for one minute. This was then washedoff with distilled water and allowed to dry. The stained smear was then observed under the microscope using oil immersion lens magnification (x100).

BIOCHEMICAL TESTS

INDOLE TEST

The test organism was inoculated into a broth that contained tryptophan and incubated at 37°C for 48 hours. Then 2ml of the broth suspension were transferred to another test tube under aseptic conditions. About 0.5ml of Kovac's reagent was added to the broth. The mixture was shaken

properly to ensure a thorough mixing and then observed for colour reaction. Positive results were indicated by a pink-coloured ring round the interface between the broth suspension and alcohol rëagent which rose to the surface.

CATALASE TEST

According to Morton, (2018). Drop a loopful of the isolate mix with the hydrogen peroxide on the slide. The production of gas bubbles (O_2) from the mixture which will occur almost immediately is a positive reaction.

RESULTS

Bacteriological analysis on deteriorated avocado pear fruit

Table 1: the summary of bacteriological examination of deteriorated avocado pear fruit.

Names of bacteria	Growth	Shape of the bacteria	Colony count	Morphology of the colony
Sample A *Shigellasp*	++	Rod	$3.9x10^4$	Smooth greyish or colorless
Sample B *Bacillus sp*	+++	Cocci	$5.1x10^5$	Rough opaque white or slightly yellow
Sample C *E.coli*	+	Rod	$1.3x10^3$	Large, thick, greyish white, moist, smooth, opaque
Sample D *Staphylococcus aureus*	++	Rod	$2.6x10^4$	Fairly large yello or white colonies.
Sample E *Salmonella sp*	+	Rod	$0.8x10^3$	Smooth colonies, strains are motile with peritrichous flagella

Note:
(+) = few growth
(++) = moderate growth
(+++) = numerous growth.
There was growth of five (5) bacteria organism that is identified. Bacillus had very much growth (5.1x105cfu/g) followed by *shigella* and *staphylococcus aureus* which was moderate growth before *salmonella sp.* which was few growth.

Table 2: Biochemical Anaysis

Reaction	Shigella	Bacillus	E. coli	Staphylococcus aureus	Salmonella
Shape	Rod	Coccus	Rod	Coccus	Rod
Gram staining	-	+	-	+	-
Catalase test	+	+	+	+	+
Indole test	+	+	+	+	+

Note:
(+) = Positive,
(-) = Negative

DISCUSSION

The bacteriological examination of the deteriorated avocado pear fruit shows that there was heavy, moderate and few growths of bacteria which is caused by thisbacteria. The bacteria utilize the amino acid and other compound of nutrient in the pear fruit in other to cause spoilage to the fruit.Avocados are a source of vitamins C, E, K, and B6, as well as riboflavin, niacin, folate,pantothenic acid, magnesium, and potassium. They also provide lutein, beta carotene, and omega-3 fatty acids. Spoilage is caused by high temperature, improperly preservation, environmental factor e.g. soil, pH and other intrinsic and extrinsic factors that has effect on the avocado pear. When eaten spoiled it tends to cause harm to the health most especially digestionalpain, stomach upset, kidney diseases and other serious tract disease.

According to Solazer*et al*., (2018) on fungi spoilage on avocado pear *Aspergillusflavus*and *Aspergillusniger* are known for their lipolytic abilities. They accelerate the breakdown of days by lipases into free fatty acid and glycerol thereby causing spoilage of oil containing fruits. A preponderance of fungi in the spoiled fruit pulp samples is because the fruit is acidic (pH 3.85). Prominent among the bacterial isolates are *Erwiniacarotovora, Pseudomonas, fluorecens, Bacillus subtilis*and*Serratiamercenccns*. The first two have been reported as causative organism for bacterial soft rots. They can like fungi hydrolyze pectin giving rise to a soft mushyappearance or consistency (Sommaruga*et al*., 2020).

The previous study shows the isolation of spoilt Avocado pear fruit like *Escherichiacoil, Proteus vulgaris, Klebsiellaaerogenes*and*Flavobacteriumsp* is suggestive of contamination from soul, harvesting equipment, handling, storage facilities and on food-contact surfaces throughout the distribution chain. Except for *Escherichia coli,* all other bacterial isolate are of no importance as agents of food borne disease. However the fruits of avocado pear should be properly processed

prior to consumption to avoid opportunistic infection (Galindo *et al*., 2021). Fresh fruits such as avocado pear are among the more challenging of food products to commercially produce and distribute. Losses due to post harvest spoilage or pathological decay are a result either of latent infections in the field that become active following harvest or of cross-contamination during harvest, cleaning, storage and distribution. Therefore spoilage management should begin in the field using an integrated strategy of good agricultural practice (GAP). It is very rich in protein, carbohydrates and minerals.

CONCLUSION

To reduce the rate of contamination which in turn leads to the spoilage of avocado pear fruits and also lead to the ingestion of contaminated fruits by consumers, it is important that the sellers should be properly educated and sensitized on the need to improve their own personal hygiene which is one of the factors that affect post-harvest of avocado pear fruits, thereby introducing contaminants and spoilage then occurs.

This research has been able to isolate and identify bacteria associated with deteriorated avocado pear fruits (*Perseaamericana*) and also present some steps to be taken in order to reduce the presence of bacteria capable of causing harm to the consumer. It is suggested that proper handling would ensure a better quality of avocado pear fruits being sold in market.

REFERENCES

Ayala, B., Silva, C., Tomas, U., Ledesma, H., and Nons, R., (2021). Avocado History, Biodiversity and Production. Sustainable Horticultural Systems. *Sustainable Development and Biodiversity*. 684(2), 157-205. doi: l0.1007/978-3-319-06904-38. ISBN 978-3-319-06903-6.

Barry, P.C. (2021). Avocado: The Early Roots of Avocado History. Canku Ota. Archived from the original source.Blumenfeld, A., Amos, K., Gazit, M., and Shmuel, V. (2014).Development of Seeded and Seedless Avocado Fruit (PDF). Agricultural Research Organization, Volcani Center, Bet Dagan, Israel.

Chen, H., Morrell, P.L., Ashworth, V., de la Cruz, M., and Clegg, M.T. (2018). Tracing the Geographic Origins of Major Avocado Cultivars.Journal of Heredity. 100 (1): 56 65. doi: 10.1093/jhered/esnO68. PMID 18779226.

Chikwe, J. (2020). Avocado market stabilizes after lockdowns. Isolates gotten from avocado pear, 678-679.

Galindo, T.M., Arzate-Fernández, A., M., Ogata-Aguilar, N., and Landero-Torres, I. (2021). The avocado (*Perseaamericana, Lauraceae*) crop in Mesoamerica (PDF). Harvard Papers in Botany. 12 (2): 325-334. doi:l0.3100/1043- 45341 2[325:TAPALC]2.0.CO;2. JSTOR 41761865.S2CID 9998040.

Lopez, G., Landon, X., Mary, G., and Amanda, J. (2019). Domestication and Significance of *Perseaamericana*, the Avocado, in Mesoamerica'. Nebraska Anthropologist. 47.

Morton, J.F. (2018). Avocado; In: Fruits of Warm Climates. Center for New Crops and Plant Products, Department of Horticulture and Landscape Architecture, Purdue University, West Lafayette, TN. pp. 91 102. ISBN 978-0-9610184-1-2.

Ohr, H.D., Coffer, M.D., and McMillan, R.T. (2023).Diseases of Avocado (*Persea Americana Miller*).*The American Phytopathological Society*.277-285.

Solazer, O., Dreher, O., Mark L., Davenport, D., and Adrienne, J. (2018). Has Avocado Composition and Potential Health Effects. *Critical Reviews in Food Science and Nutrition*. 53 (7): 738 750. doi:l0.1080/10408398.2011.556759. ISSN 1040-8398.PMC 3664913.PMID 23638933.

Sommaruga, R., Eldridge, B., and Honor, M., (2020). Avocado production: Water footprint and socioeconomic implications. EuroChoices. 20 (2): 48 53. doi:10.1 11 l/1746-692x.12289. ISSN 1478-09 17.S2CID 230594487.