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Microbial and physiochemical quality of post harvest apples (*Malus domestica* Borkh) influenced by storage condition

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ABSTRACT

This study assess the microbial and physiochemical quality of post-harvested apples (Malus domestica) under varying storage conditions, specifically ambient temperature and refrigeration temperature. Microbial analyses revealed distinct trends in bacterial and fungal counts. Standard microbiological and physiochemical techniques were adopted. For refrigerated apples, the total heterotrophic bacteria count peaked on Day 15 (1.8×10^5 CFU/g), contrasting with the lowest count on Day 3 (3.6×10³ CFU/g). Similarly, ambient temperature of storage exhibited highest aerobic bacteria growth on Day 15 $(3.3 \times 10^4 \text{ CFU/g})$ and lowest on Day 2 $(5.0 \times 10^3 \text{ CFU/g})$. Staphylococcus counts in refrigerated apples peaked on Day 3 (3.6×10³ CFU/g) and ambient temperature storage on Day 15 $(7.4 \times 10^3 \text{ CFU/g})$. The study identified seven bacterial genera, including *Staphylococcus* (19.6%), Bacillus (21.6%), and Proteus (17.6%) being prominent. Others were; 11.8% Micrococcus; 21.6%, Corynebacterium,; 13.7% Pseudomonas and 5.9% Citrobacter. Physiochemical attributes exhibited temperature-dependent variations. pH levels in refrigerated apples were lowest on Day 15 (2.88) and highest on Day 12 (3.80), whereas ambient temperature storage showed the highest pH (3.59) on Day 0 and lowest on Day 12 (3.42). Moisture content ranged from 80.2% (Day 3) to 87.8% (Day 15) for refrigerated apples, and from 85.7% (Day 1) to 87.8% (Day 15) for ambient temperature storage. Specific gravity decreased from 0.64 (Day 0) to 0.51 (Day 15) for refrigerated apples and from 0.64 (Day 0) to 0.59 (Day 15) for ambient temperature storage. Total suspended solids (TSS) fluctuated from 17.86% (Day 0) to 12.01% (Day 15) for ambient temperature and 12.54% (Day 15) for refrigerated temperature. The study recommends implementing optimized refrigeration protocols, considering the observed microbial growth patterns, could enhance the preservation of apples. While emphasizing stringent hygiene practices within storage facilities to be crucial, regular cleaning and disinfection routines even in refrigerators is a necessity.

Keywords: Post harvest apple, Ambient temperature, Refrigeration temperature microbial and physiochemical quality

1. INTRODUCTION

The demand for fresh fruit increases by more than 10% annually. Increasing production rate does not ensure a stable and uniform supply of fresh products to the consumer. Solving the problem requires not only equipping the horticulture industry with modern equipment, but also the use of advanced fruit storage technologies. The storability (shelf life) is determined by the ability of fruits to remain for a certain time without significant weight loss and degradation of commercial qualities (Kopylov et al., 2017). The quality and storability of fruit batches are formed under the influence of a complex of environmental, biological, agrotechnical factors that can act as sources and activators of fruit growth and development, as well as triggers for the development of physiological diseases and injuries, or contribute to an increase in the degree of manifestation of damages. Diseases and damages "acquired" by fruits during the pre-harvest period are most often intensified during storage, resulting in losses of up to 30% or more. The key factors determining the vital activity of fruits in the period after harvesting are temperature (T°), as well as the content of oxygen (O₂) and carbon dioxide (CO₂) and relative humidity (%) in the storage atmosphere (Plugatar et al., 2021).

Apple (*Malus domestica*) is one of the highly ephemeral and important fruits of the world, mainly grown in temperate regions. It belongs to the family of Rosaceae and sub-family Pomoideae (Spengler, 2019). More than 7500 apple cultivars are known, but only a few of them are commercially important. Some varieties like Red Delicious, Golden Delicious, Granny Smith, Rome Beauty and McIntosh are commonly produced worldwide. A low percentage of apples produced are consumed immediately after harvesting, and most of the time, a large part of them is stored for a long period to keep them available for their further utilization (Grabska et al., 2023).

Plants became the basis of traditional medicine system throughout the world for thousands of years and continue to provide mankind with new remedies. Two terms namely, quality and acceptability play an important role in the selection of fruits like apples. The term quality can be defined as the combination of characteristics of fruit and the consumer's recognition. On the other hand, the feedback to those characteristics is referred to as acceptability (Yousuf et al., 2020). The quality of most of the fruits varies with time as well as the quality of the individual fruit may also differ extremely from the average. For example, apple is one of those fruits whose quality changes rapidly with time during the storage period, due to which its acceptability among consumers also varies (Viera et al., 2009). The quality of an apple or any fruit can be predicted by various properties or attributes like sensory, physical, chemical, mechanical, microbial, physicochemical etc. In the first instance, it is judged by its appearance, including size, gloss, and color, and thereafter by its firmness, density, acidity, and total soluble solids content (Jha et al., 2012). These physicochemical attributes may help consumers to recognize the nutritional value of fruits (Jha et al., 2012). Consumers do not like light weight, colorless, and shriveled fruits. In the past several years, numerous studies have been conducted to improve the quality of apples during storage using various treatments (Ali et al., 2018).

Moreover, consumers generally purchase apples on the basis of their appearance, only because at that time, their other quality parameters such as taste, acidity, total soluble solids content, etc. are not accessible.

Reduction in quality of apple occur mainly during handling and storage. Poor storage of fruits allow microbial silage to occur and can contribute significantly to lowering the quality of fruits. The microorganisms implicated in the spoilage of apples could be from rough and unhygienic handling or from soil, dust, wind etc. Microorganisms from these sources can be deposited on the surface of the fruit and can vary from not only one orchard to the next, but from fruits to fruits. The study is aimed at determining the microbial and physiochemical quality of post harvest apple.

2. MATERIALS AND METHODS

Study area / sample collection

Fifteen (15) apple samples were purchased from the same vendor at Choba market in Obio-Akpor Local Government Area, of Rivers State to reflect similar post-harvest and storage duration. The samples were placed in a sterilized bag and transported to the Microbiology laboratory for analysis. Microbial analysis was done on days 0, 3, 6, 9, 12, and 15 of storage while physiochemical analysis was done on days 0, 3, 6, 9 and 15 respectively.

Processing of Samples

All Apple samples purchased from the market were bought and categorized into two groups, one was refrigerated and the other kept at ambient temperature. One apple was withdrawn from each group on each day of analysis.

Bacteriological Analysis

25g of each apple sample was weighed into a stomacher bag containing 225 ml of sterile diluent (peptone water), homogenized in a stomacher for 2 mins to obtain the stock solution. Ten-fold serial dilution was performed on the samples. 1 ml of the aliquot was pipetted into a test-tube containing 9 ml sterile 0.1% peptone water to make 10^{-1} , 10^{-2} and 10^{-3} . Using a sterile 1 ml pipette (syringe), 1 ml of each of the dilution were inoculated on the different agar plates for enumeration and culture .

Enumeration of microbial count/Purification of Isolates

The microbial count for each sample was obtained on different agars including Plate Count Agar, MacConkey agar and Mannitol Salt agar. Pure culture was then gotten from the previously incubated Petri dishes and counts obtained from the various selective media were expressed as a colony forming unit (cfu/g). Colonies were sub-cultured on nutrient agar by streak plate method to obtain pure distinct colony.

Total Heterotrophic Bacteria Count

1 ml of diluted sample was added onto the solidified PCA plates and spread on the surface using a sterile glass rod. The plates were then incubated at 37 °C for 24 hours. Colonies observed were enumerated and sub-cultured on nutrient agar plates to purify and stored on agar slants.

Total Staphylococcus Counts

1 ml of diluted sample was added onto the solidified MSA plates and spread on the surface using a sterile glass rod. The plates were then incubated at 37 °C for 24 hours. Colonies observed were enumerated and sub-cultured on nutrient agar plates to purify and stored on agar slants.

Total Coliform Counts

1 ml of diluted sample was added onto the solidified MacConkey plates and spread on the surface using a sterile glass rod. The plates were then incubated at 37 °C for 24 hours. Colonies observed were enumerated and sub-cultured on nutrient agar plates to purify and stored on agar slants.

Purification and Storage of Bacterial Isolates

Streak plating technique was done on nutrient agar plates using for all isolates obtained from the various selective media. The nutrient agar was prepared by dissolving 28 g of nutrient agar powder in 1 L of distilled water, mixed properly and autoclaved for 121 °C for 15 min at 15 psi, allowed to cool a little before dispensing into sterile Petri dishes and allowed to solidify. Distinct colonies from the differential agar plates were picked and streaked on the solidified nutrient agar medium and incubated for 24 hrs at 37 °C to purify. Purified distinct colony were then picked and introduced into already prepared nutried agar slants and stored in refrigerator until reuse. The identities of the isolates was confirmed using biochemical tests (Cheesebrough, 2005). Tests carried out include indole, catalase, methyl red production, Voges-Proskauer reaction, citrate and triple sugar iron agar test (TSIA) and sugar fermentation tests for all isolates.

Examination of fungi

The cultural characteristics of each fungi isolates was identified according to their colour, shape and the cell morphology was done based on mycelia, hyphae, septate, spore formation using lactophenol blue. A piece of the mycelium from the Petri plates was mounted on a clean grease free slide using a sterile wire loop and covered with a cover slip, after which a drop of lactophenol cotton blue was added and examined with the microscope.

Physiochemical Analysis

Determination of pH

The pH of each group of apple sample stored at the different temperature determined with a pH meter. The juice was extracted and ten milliliter of the juice was dispensed into a beaker and the pH was determined with a previously standardized pH meter as described by Ahmed et al., (2021).

Determination of Moisture Content

One gram of the sample was weighed into a clean dried porcelain evaporating dish. This was placed in an oven to maintain a temperature of 105 °C for 6 hours. The evaporating dish was cooled in desiccator to room temperature, then it was re-weighed and recorded as described by Ahmed et al., (2021).

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 $Moisture = \frac{Weight of fresh sample - Weight of dried sample}{Weight of sample used} \times 100$

Determination of Specific Gravity

Specific gravity was determined by water displaced method as described by Mazumdar & Majumder (2003). Apple fruit were washed with distilled water, dried and weighed one by one. Measuring cylinder with a capacity of 1000 ml was taken and filled with 500 ml of distilled water. Apple fruits were dipped in measuring cylinder. The volume of water displaced was noted as decribed by by Khan et al., (2017).

Specific gravity = $\frac{\text{Weight of apple fruit}}{\text{Volume of water displaced}}$

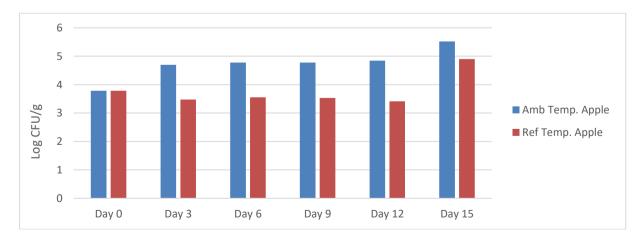
Determination of Total Soluble Solid

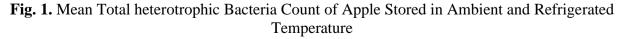
Total Soluble Solids (%) TSS were determined using Atago RX 1000 digital refractometer. A drop of juice was extracted and placed on clean prism of Refractometer and the lid was closed. Reading was taken directly from the scale at room temperature as described by Khan et al., (2017)

3. RESULTS AND DISCUSSION

Total heterotrophic Bacteria Count of Apple Stored in Ambient and Refrigerated Temperature

The total heterotrophic bacteria count of apples stored at different temperature showed that the highest count was observed on Day 15 ($8.0 \times 10^4 \text{ CFU/g}$) for the refrigerated apples and the least was observed on day 12 ($2.6 \times 10^3 \text{ CFU/g}$). The apples stored at ambient temperature had highest aerobic bacteria growth on day 15 ($3.3 \times 10^5 \text{ CFU/g}$) while the least was observed on day 1 ($6.1 \times 10^3 \text{ CFU/g}$).





Total Staphylococcus Bacteria Count of Apple Stored in Ambient and Refrigerated Temperature

The total staphylococcus count of apples stored at different temperature showed that the highest count was observed on Day 3 $(3.6 \times 10^3 \text{ CFU/g})$ for the refrigerated apples and the least was observed on day 1 $(4.5 \times 10^1 \text{ CFU/g})$. The apples stored at ambient temperature had Staphylococcus count on day 15 $(7.4 \times 10^3 \text{ CFU/g})$ while the least was observed on day 1 $(4.5 \times 10^1 \text{ CFU/g})$.

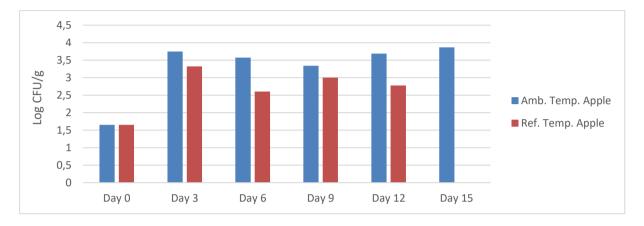
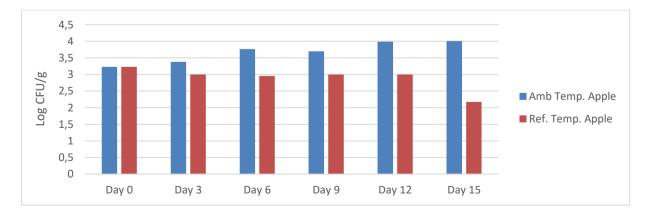
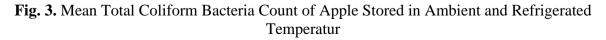


Fig. 2. Mean Total Staphylococcus Bacteria Count of Apple Stored in Ambient and Refrigerated Temperature

Total Coliform Bacteria Count of Apple Stored in Ambient and Refrigerated Temperature

The total coliform count of apples stored at different temperature showed that the highest count was observed on Day 1 $(1.7 \times 10^3 \text{ CFU/g})$ for the refrigerated apples and the least was observed on day 15 $(1.5 \times 10^2 \text{ CFU/g})$. The apples stored at ambient temperature had highest aerobic bacteria growth on day 15 $(1.02 \times 10^4 \text{ CFU/g})$ while the least was observed on day 1 $(1.7 \times 10^3 \text{ CFU/g})$.





Total Fungi Count of Apple Stored in Ambient and Refrigerated Temperature

The total fungi count of apples stored at different temperature showed that the highest count was observed on Day 6 $(4.0 \times 10^2 \text{ CFU/g})$ for the refrigerated apples and the least was observed on day 3 $(4.0 \times 10^2 \text{ CFU/g})$. The apples stored at ambient temperature had highest fungal growth on day 3 $(2.8 \times 10^3 \text{ CFU/g})$ while the least was observed on day 1 $(5.0 \times 10^2 \text{ CFU/g})$.

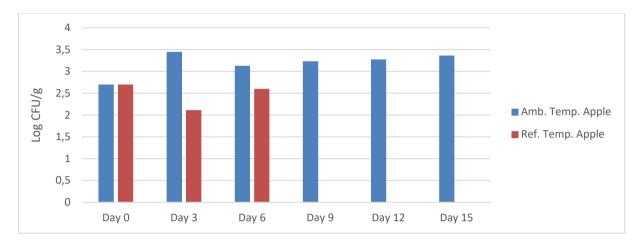


Fig. 4. Mean Total Fungi Count of Apple Stored in Ambient and Refrigerated Temperature

Frequency of Occurrence of Bacteria Isolated from Apple Stored in Ambient and Refrigerated Temperature

The frequency of occurrence of the different isolates from the different storage conditions studied. Table 1 shows the distribution of isolates among the species studied. Fig. 4 shows a pie chat of the overall frequency of occurrence of all isolates. The frequencies were 19.6% *Staphylococcus*, 11.8% *Micrococcus*, 21.6% *Bacillus*, 8% *Corynebacterium*, 13.7% *Pseudomonas*, 5.9% *Citrobacter* and 17.6% *Proteus*.

Bacteria genera	Ambient temperature n (%)	Refrigerated temperature n (%)	Total N (%)	
Micrococcus spp.	5 (16.1)	1(5.0)	6 (11.8)	
Staphylococcus spp.	5 (16.1)	5(25.0)	10(19.6)	
Bacillus spp.	8 (25.8)	3 (15.0)	11(21.6)	
Corynebacterium spp.	4 (12.9)	1(1.0%)	5(9.8)	
Pseudomonas spp.	3 (9.8)	4(20.0)	7(13.7)	

Table 1. Frequency of Occurrence of Bacteria Isolated from Different Study Conditions

Citrobacter spp.	1(3.2)	2(10.0)	3(5.9)	
Proteus spp.	5(16.1)	4(20.0)	9(17.6)	
Total	31 (100)	20 (100)	51 (100)	

Physiochemical Attribute of Apple stored at different Temperature

The physiochemical attribute of apple stored at different temperature varied. The pH was lowest for day 15 (2.88) for the refrigerated and highest on day 12 (3.80). Moisture content was highest for 87.8% on day 15 and least 80.2% on day 3. The apple stored at ambient temperature had highest pH value of 3.59 on day 0 and the least on day 12 (3.42). The moisture content was 85.7% on first day while the last day (day 15) was 87.8 for ambient temperature and 87.8 for refrigerated apples. The specific gravity was 0.64 on day 0 while the last day was 0.51 and 0.59 apples stored at room and refrigerated temperatures. The total suspended solid (TSS) waS 17.86% on day 0 while the 15th day had TSS of 12.01% for ambient temperature and 12.54% for refrigerated temperature.

Days	Ambient Temperature			Refrigerated Temperature				
	рН	Moisture (%)	Specific Gravity	TSS (%)	pН	Moisture (%)	Specific Gravity	TSS (%)
Day 0	3.59	85.7	0.64	17.86	3.59	85.7	0.64	17.86
Day 3	3.52	80.2	0.61	15.22	3.45	80.2	0.64	15.32
Day 6	3.44	87.0	0.61	15.74	3.69	87.0	0.63	16.82
Day 9	3.43	87.5	0.58	15.74	3.78	87.5	0.62	16.43
Day 12	3.42	87.6	0.55	13.52	3.80	87.6	0.61	15.83
Day 15	3.40	87.8	0.51	12.01	2.88	87.8	0.59	15.54

Table 2. Physiochemical Attribute of Apple stored at different Temperature

4. DISCUSSION

Microbial Quality of Apple Stored at Different Storage Temperature

Apple is one of the frequently eaten fruits in this part of the world. Due to its nutritional values, people tend to store remnants of the fruit in refrigerators to avoid wastage in a world faced with food security challenges (Wani, et al., 2023; Karanth et al., 2023). Hence, this study investigated the impact of storage conditions, specifically ambient temperature and refrigerated temperature, on the microbial and physiochemical quality of post-harvested apples (*Malus domestica*). By examining the dynamic interplay between storage environments and the

microbial composition, as well as physiochemical attributes, this research aims to enhance our understanding of optimal storage practices for preserving the overall quality and safety of harvested apples.

The results indicate that the total heterotrophic bacteria count varied significantly with storage temperature. For refrigerated apples, the highest count was observed on Day 15 $(8.0 \times 10^4 \text{ cfu/g})$, while the least count was noted on Day 12 (2.6×10³ cfu/g). In contrast, apples stored at ambient temperature exhibited the highest aerobic bacteria growth on Day 15 (3.3×10^5) cfu/g), with the least count observed on Day 1 (6.1×10³ cfu/g). These findings suggest that refrigeration tends to delay the growth of heterotrophic bacteria compared to ambient conditions. The heterotrophic bacteria count ranges obtained in this study showed that both fruits are still fit for consumption as results qualify the apples studied as "not spoil" and "fit" for human consumption. Hazard Analysis and Critical Control Point-Total Quality Management (HACCP-TOM) Technical Guidelines rates microbial quality for raw foods containing aerobic plate count of 5×10⁷ CFU/g "Spoilt" (EC-SCF, 2002; Aycicek et al., 2006). Similarly, the total staphylococcus count displayed a similar trend, with refrigerated apples reaching their peak on Day 3 (3.6×10^3 cfu/g) and the lowest count on day 1 (4.5×10^1 cfu/g). in the case of ambient-stored apples, the highest staphylococcus count was observed on day 15 $(7.4 \times 10^3 \text{ cfu/g})$, and the lowest was on day 1 $(4.5 \times 10^1 \text{ cfu/g})$. these findings suggest that refrigeration may slow down the proliferation of Staphylococcus on apple surfaces.

The total coliform count exhibited different patterns, with refrigerated apples reaching the highest count on Day 1 $(1.7 \times 10^3 \text{ cfu/g})$ and the lowest on day 15 $(1.5 \times 10^2 \text{ cfu/g})$. in contrast, apples stored at ambient temperature displayed the highest coliform count on day 15 $(1.02 \times 10^4 \text{ cfu/g})$ and the lowest on day 1 $(1.7 \times 10^3 \text{ cfu/g})$. this indicates that coliform growth may be affected differently by storage temperatures, with refrigeration showing an initial increase followed by a decline.

The coliform counts reported in this work is lower than the report of De Giusti et al. (2010), it is however also lower than the findings of Viswanathan and Kaur (2001) and it is in agreement with the reports of Aycicek et al. (2006), Bagci and Temiz (2011), Ukwo et al. (2011), Mahuya et al. (2011) and Joceyln et al. (2012). Coliforms are indicator organisms and counts of $10^4 - 10^6$ cfu/sample reported in this work are a cause for concern, since the fruits are usually consumed without further processing. In general, the microbial Counts obtained in this study is comparable with the findings of Oranusi and Wesly (2012) who reported a count of $3.4 \times 10^5 - 4.5 \times 10^5$ cfu/g was obtained for Heterotrophic bacteria, while total coliform and total fungal counts ranges from 2.4×10^4 to 2.2×10^6 and 5.0×10^2 to 3.6×10^5 cfu/mL respectively during a study of apple fruits sold in Owerri.

The total fungi count also varied, with refrigerated apples showing the highest count on Day 3 $(1.3 \times 10^3 \text{ CFU/g})$ and the least on Day 6 $(4.0 \times 10^2 \text{ CFU/g})$. Ambient temperature-stored apples exhibited the highest fungal growth on Day 15 $(2.3 \times 10^3 \text{ CFU/g})$ and the least on Day 1 $(5.0 \times 10^2 \text{ CFU/g})$. These results suggest that refrigeration may inhibit fungal growth to some extent. Fungal growth varied, with refrigerated apples peaking on Day 3 $(1.3 \times 10^3 \text{ CFU/g})$ and ambient temperature storage showing a peak on Day 15 $(2.3 \times 10^3 \text{ CFU/g})$.

The identified bacterial genera, including *Staphylococcus, Micrococcus, Bacillus, Corynebacterium, Pseudomonas, Citrobacter,* and *Proteus,* displayed differing prevalence across storage conditions, indicating temperature-dependent microbial dynamics. The microorganisms isolated in this work corroborate the findings of other studies on fruits and vegetables (Abadias et al., 2008; Uzeh et al., 2009; Bucker et al., 2010; Eni et al., 2010; Ukwo

et al., 2011; Joceyln et al., 2012). The frequency of occurrence of these isolates varied, with Staphylococcus being the most prevalent 19.6% However frequency was 11.8% *Micrococcus*, 21.6% *Bacillus*, 9.8% *Corynebacterium*, 13.7% *Pseudomonas*, 5.9% *Citrobacter* and 17.6% *Proteus*. Comparing our results with microbial standards set by WHO (1.0×10^4 CFU/g), it is evident that refrigerated storage aligns better with recommended practices to minimize microbial contamination.

In generally, the study revealed that in most cases, the microbial load of apples stored at refrigerated temperature had higher counts than the one stored at ambient temperature, these could be due to several reasons because in general, storing apples at refrigerated temperatures is expected to inhibit the growth of microbes and slow down the spoilage process compared to storing them at ambient temperature. Refrigeration helps to slow down the metabolic activities of microorganisms, including bacteria and fungi, reducing their growth rate and extending the shelf life of fruits. However, the reason could be contamination during handling. If apples are not properly washed or handled before being placed in the refrigerator, they may carry a higher microbial load into the cold storage. Contaminants from hands, surfaces, or other sources can introduce microorganisms.

Also, Refrigerators can sometimes create a humid environment, especially if the apples are not stored in breathable containers or perforated bags. High humidity can create conditions favorable for the growth of certain microbes, potentially leading to an increase in microbial load .If apples are damaged or bruised, whether before or during refrigeration, it can create entry points for microbes to colonize and grow. Refrigeration may slow down their growth, but it won't necessarily eliminate existing microbial populations on damaged areas. Microbes from one food item can transfer to another, including the apples, increasing the overall microbial load. The inconsistency in power supply is also implicated to the dwindling result recorded as power outage is a challenge and refrigeration temperature fluctuates between ambient and room temperature almost on a daily basis..

Some of the bacterial isolates from apple fruits are Gram negatives and non pathogenic, however, the presence *E. coli, Salmonella* spp, *Shigella* spp which are often associated with poor sanitary practices indicate that they put a pointer to a potential risk of food borne illness to consumers (Aycicek et al., 2006; Oranusi et al., 2006, 2007; Eni et al., 2010). *Staphylococcus aureus* and *B. cereus* are common food contaminants from Man and the environment, their presence in food however, need to be controlled because they have been reported as cause of major food borne illnesses (Mudgil et al., 2004; Oranusi et al., 2004, 2006a, 2006b, 2007). The fungal isolates of apple fruits in this study *Aspergillus* spp; *Penicillium* spp, *Rhizopus, Mucor* spp are common environmental contaminants, they have been reported by other researchers (*Tournas*, 2005; *Badosa* et al., 2008). They are known to be the major cause of spoilage of fruits and vegetables (ICMSF, 1998). Some of these fungi have been reported to produce mycotoxins and are implicated in cases of mycoses (Tournas, 2005; Katherine et al., 2006)

Raw fruits and vegetables are known potential for a wide range of microorganisms, including human pathogens (EC-SCF, 2002). The survival or growth of these organisms on intact fruit surfaces will be dependent on the extrinsic factors of available nutrient, temperature, presence of scales and fibres, gaseous atmosphere, mechanical handling and moisture. The apple fruits on display for sale are often visited by many hands of the customers and by the vendors. These individuals pick and drop as many apple fruits as are available, to enable them make a choice. Poor handling by unhygienic hands is a factor contributing to the high microbial load.

The dusty environments of the motor parks, busy roads and campuses/institutions, coupled with water of questionable quality which often is used to sprinkle the fruits to keep fresh are contributing factors that could aid the survival and possible multiplication of contaminants on fruit surfaces.

Physiochemical Attributes of Apple Stored at Different Temperature

The pH levels of the apples exhibited dynamic changes throughout the storage period. Refrigerated apples showed the lowest pH on Day 15 (2.88), whereas ambient temperature storage displayed the highest pH on Day 0 (3.59). The mean pH for ambient temperature is 3.46 and that of the refrigeration temperature is 3.53 the values recorded in our study is comparable to Khan et al., (2017), Who reported the pH of stored apples increased at refrigeration temperatures of 7 °C (4.53) and decreased at cold storage temperature of 16 °C (4.20). They also observed that extending storage periods resulted in increase in pH from 0 to 14 days in their study. These variations in pH suggest that refrigeration may have a distinct impact on the acidity of apples, potentially influencing factors such as taste, texture, and susceptibility to microbial growth. The observed increase in pH for ambient-stored apples on Day 0 could be attributed to initial microbial activity or other biochemical processes.

Moisture content is a critical parameter affecting the texture, juiciness, and overall quality of apples. For refrigerated apples, moisture content ranged from 80.2% (Day 3) to 87.8% (Day 15), while for ambient-stored apples, it fluctuated between 85.7% (Day 1) and 87.8% (Day 15). The differences in moisture content suggest that refrigeration may contribute to maintaining a more consistent moisture level over the storage period compared to ambient conditions. This is crucial for preventing dehydration or excessive water loss, which can affect the sensory attributes of the fruit.

Specific gravity, indicative of apple density and internal structure, experienced a notable decrease during the storage period for both refrigerated and ambient-stored apples. For refrigerated apples, specific gravity decreased from 0.64 (Day 0) to 0.51 (Day 15), and for ambient-stored apples, it decreased from 0.64 (Day 0) to 0.59 (Day 15). Our result can be compared to Khan et al., (2017) who report a specific gravity of 0.62 and 0.62 for refrigerated and ambient temperature stored apples. These changes may be associated with alterations in cell structure, water content, or other physiochemical transformations occurring during storage.

The observed differences between the two storage conditions emphasize the need to consider specific gravity as an essential parameter in assessing apple quality under different storage temperatures.

Total Soluble Solid, representing the concentration of dissolved solids in the fruit, fluctuated during the storage period. On Day 15, ambient-stored apples exhibited a TSS value of 12.01%, while refrigerated apples showed a slightly higher value of 12.54%. Values obtained in this study is comparable wutj Ahmad et al., (2021) who reported similar ranges (11.88 to 7.68) in TSS in their study. These findings indicate that refrigeration may impact the accumulation of soluble solids in apples, potentially influencing factors such as sweetness and flavor. The intricate relationship between storage temperatures and the microbial-physiochemical quality of post-harvested apples has been elucidated through this study. The observed variations in pH levels, moisture content, specific gravity, and TSS provide valuable insights for developing effective storage and preservation strategies in the fruit industry. Understanding how different storage conditions influence these parameters is essential for optimizing apple quality and shelf life, ultimately benefiting both producers and consumers in

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the fruit supply chain. This research into the microbial and physiochemical quality of postharvested apples (Malus domestica) under varying storage conditions has yielded significant insights into the intricate dynamics that influence the overall quality and safety of stored apples.

The culmination of extensive analyses provides a nuanced understanding of the microbial composition and physiochemical attributes, emphasizing the critical role of storage conditions in shaping the fate of harvested apples. The study revealed substantial variations in microbial populations based on storage conditions and duration. Refrigerated apples exhibited the highest heterotrophic bacteria count on Day 15, highlighting the impact of prolonged cold storage in mitigating bacterial proliferation. Conversely, ambient temperature storage witnessed rapid aerobic bacteria growth on Day 15, indicating a need for vigilant monitoring during extended storage periods.

Physiochemical attributes exhibited temperature-dependent fluctuations, impacting the overall quality of stored apples. The variation in pH levels and moisture content indicated the sensitivity of apples to storage conditions, with potential implications for taste, texture, and overall consumer acceptance. Specific gravity and total suspended solids (TSS) fluctuated, further highlighting the complex interplay between storage conditions and the physical attributes of apples.

5. CONCLUSION

This study not only advances our understanding of the microbial and physiochemical aspects of stored apples but also serves as a foundation for continuous improvement in postharvest management strategies, contributing to the enhancement of food safety and quality in the fruit industry.

Recommendations

Implementing optimized refrigeration protocols, considering the observed microbial growth patterns, could enhance the preservation of apples. Fine-tuning temperature and humidity controls, especially during prolonged storage, may contribute to a reduction in bacterial and fungal proliferation

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