



# The Effects of Different Storage Temperatures on the Microbial, Physicochemical and Organoleptic Quality Changes in the Shellfish "Ngolo" (*Thais califera*) from Nigeria

Boma O. Oruwari<sup>1</sup> and Bernard J. O. Efiuwewere<sup>2\*</sup>

<sup>1</sup>Research and Development Division, Nigerian National Petroleum Corporation (NNPC),  
4-9 Moscow Road, Port Harcourt, Nigeria.

<sup>2</sup>Department of Microbiology (Food & Industrial Division), University of Port Harcourt, Port Harcourt,  
Nigeria.

## Authors' contributions

This investigation was carried out in collaboration between both authors. Author BJOE designed the study and supervised the work. Author BOO wrote the first draft of the manuscript and managed the literature searches and analysis of the data. Both authors read and approved the final manuscript.

## Article Information

DOI: 10.9734/BMRJ/2016/27698

### Editor(s):

(1) Lachhman Das Singla, Department of Veterinary Parasitology, College of Veterinary Science, Guru Angad Dev Veterinary and Animal Sciences University, India.

### Reviewers:

(1) Francesca Piras, University of Sassari, Italy.

(2) Alex Augusto Gonçalves, Federal Rural University of Semi Arid, Brazil.

Complete Peer review History: <http://www.sciencedomain.org/review-history/15666>

Original Research Article

Received 14<sup>th</sup> June 2016  
Accepted 23<sup>rd</sup> July 2016  
Published 5<sup>th</sup> August 2016

## ABSTRACT

**Aims:** To investigate the effects of different storage temperatures ( $-15\pm 2^\circ\text{C}$ ;  $4\pm 2^\circ\text{C}$  and  $29\pm 2^\circ\text{C}$ ) on quality changes in 'Ngolo' (*Thais califera*) to evaluate the potential enhancement or otherwise of its domestic and global trade.

**Study Design:** "Ngolo" samples were subjected to different temperatures and evaluated for microbiological, physico-chemical and organoleptic changes during short and long-term storage.

**Place and Duration of Study:** Department of Microbiology (Food and Industrial Division), University of Port Harcourt and Research and Development Division, Nigerian National Petroleum Corporation (NNPC), Moscow Road, Port Harcourt, Nigeria between January 2006 and December 2008.

\*Corresponding author: E-mail: [bjefiuvv@yahoo.com](mailto:bjefiuvv@yahoo.com);

**Methodology:** Freshly shucked "Ngolo" samples were divided using sterile polyethylene bags into sub-samples of 150 g per bag and stored at (-15±2°C; 4±2°C and 29±2°C) respectively and analysed at daily or weekly intervals for microbial, physico-chemical and organoleptic quality attributes.

**Results:** Variations in quality attributes of "Ngolo" samples were induced by the temperatures resulting in significant ( $P=0.05$ ) minimum total viable counts (2.12 log CFU/g) in samples stored at -15±2°C compared with 8.72 log CFU/g in samples stored at 29±2°C. Little or no change in pH occurred within two weeks in samples stored at -15±2°C and 4±2°C respectively but decrease in pH within 24h occurred in samples stored at 29±2°C. Total volatile nitrogen (TVN) and trimethylamine (TMA) contents remained low (32 mgN/100 g TVN and 25 mgN/100 g TMA) in samples stored at -15±2°C and were acceptable throughout storage while the contents of those stored at 29±2°C increased significantly ( $P=0.05$ ) from day 0 to 72 mgN/100 g and 48 mgN/100g respectively on day 4 and were rejected within 24h. Significant ( $P=0.05$ ) positive correlations between TVCs and TVN occurred indicating their usefulness for prediction of quality changes in "Ngolo" (and could be applied to other related seafoods).

**Conclusion:** Overall, the quality attributes of samples stored at -15±2°C remained significantly ( $P=0.05$ ) acceptable throughout storage thereby enhancing their potential for international food trade.

*Keywords: Molluscan (Thais califera); shellfish; microbial safety; temperature effects.*

## 1. INTRODUCTION

Shellfish are economically important seafoods in various parts of the world including the Niger Delta Region of Nigeria. They constitute a major source of protein supply worldwide and are readily available in the Niger Delta Region of Nigeria [1,2,3].

"Ngolo" (*Thais califera*) is a univalve molluscan shellfish, commonly called whelk, occurring in the marine and brackish waters of the West African Coast and elsewhere [1,4]. They remain in good condition if un-shucked but once shucked, they deteriorate except they are subjected to preservative treatments. Spoilage of shellfish is mainly due to bacterial activities which lead to several quality changes including off-odour, off-flavour and adverse colour changes [5,6]. The most common method of preserving shellfish in the fresh form is by freezing or low temperature storage [5,6,7].

The microbiological and spoilage characteristics of shellfish from temperate waters have received much attention [5,7,8]. In contrast, little or no research work has been reported on shellfish (including "Ngolo") obtained from the Niger Delta region of Nigeria [1,3], and other parts of the world [4,9] in spite of their abundance and economic importance in these regions.

In view of the high perishability of "Ngolo" (*Thais califera*) as well as the interest in shellfish trade globalization, the present investigation was therefore undertaken to determine the

microbiological, physicochemical and organoleptic changes in this important shellfish during storage under different temperatures commonly encountered in developing and developed countries for commercial enterprises.

## 2. MATERIALS AND METHODS

### 2.1 Sample Collection and Processing

"Ngolo" (*Thais califera*) samples were harvested manually around Idama village along the Sombreiro River, Rivers State, Nigeria and collected in pre-sterilized polyethylene bags, sealed and transported in ice coolers to the laboratory for analyses [9].

Shucking of the "Ngolo" meat from the shell was carried out aseptically by sterilizing the surface of the "Ngolo" shell with 70% ethanol and cracking the "Ngolo" shell with a small pre-sterilized hammer on an improvised pre-sterilized anvil. The "Ngolo" flesh was then removed from the cracked shells with the aid of sterile forceps.

### 2.2 Storage and Analysis of Samples

The shucked "Ngolo" samples were aseptically divided into sterile polyethylene bags (150g per bag) and stored at -15±2°C (in Haier Thermocool Ultra Low temperature Freezer; model DW86L388, Qingdao, China); 4±2°C and 29±2°C respectively. Sets of samples stored at 29±2°C were analysed for quality changes on day 0 and daily for four days while "Ngolo" samples stored at -15±2°C (thawed at about

30°C for 2.5h); 4±2°C were analyzed on weekly intervals for 12 weeks and daily for 4 days respectively. The parameters evaluated during the storage were: Total viable counts (TVCs), physicochemical [(pH, Total Volatile Nitrogen (TVN), Trimethylamine (TMA)] and organoleptic attributes (visual appearance, flavour, texture and taste).

### 2.3 Microbiological Analysis of Samples

The spread-plate technique was adopted for the enumeration of the TVCs of the "Ngolo" samples stored at the different temperatures [10].

The shucked "Ngolo" meat (50 g) was aseptically weighed and homogenized in a pre-sterilized blender (Moulinex, Paris, France) containing 450 ml of 0.1% (w/v) sterile peptone water to obtain a 1:10 dilution. Further decimal dilutions of 10<sup>-2</sup> to 10<sup>-7</sup> of the homogenate were prepared and plated on pre-dried tryptone soy-agar (Biotec, Suffolk, United Kingdom) plates in duplicate. The inoculated plates were incubated at 37°C for 24h and the developed colonies (30-300) were enumerated.

Representative bacterial colonies were isolated, characterized presumptively by colonial morphology, motility, pigmentation and Gram staining before biochemical tests were carried out. The biochemical tests for the identification of the bacterial isolates included the following criteria: indole, methyl red, Voges-Proskauer, citrate utilization (IMViC) tests, fermentation of glucose, fructose, sucrose, mannitol, maltose, arabinose and lactose as well as catalase, oxidase and coagulase tests based on previously described methods [10,11].

### 2.4 Physicochemical Analysis of Samples

The physicochemical (pH, TVN and TMA) tests were carried out on the stored "Ngolo" samples following earlier descriptions of Pearson [12]. The pH was determined by homogenizing the "Ngolo" meat samples in deionised water (1:2 ratio) using pre-sterilized blender (Moulinex, Paris, France) and then measured using pH meter (Hach Sension I, Loveland, USA) after it had been calibrated with buffer solutions of 4.0 and 7.0 [13]. The semi-micro-steam distillation method was used in determining the TVN and TMA contents of the samples [12]. The slurry

was centrifuged at 1800 rev. min<sup>-1</sup> (Uniscopce Lab. Centrifuge, Model SM112, England). Extracts from the homogenized and centrifuged samples were distilled with 2M NaOH and formaldehyde and the distillate titrated against 0.01M NaOH and the TMA and TVN determined [12] three replicates in duplicate using a spectrophotometer (model SP-3000 PLUS, Optima Inc., Tokyo, Japan).

### 2.5 Determination of Sensory Quality Attributes of Samples

"Ngolo" samples taken from the different storage temperature were boiled for 10 min and evaluated by a 10- member taste panel consisting of staff and others who are very conversant and knowledgeable about the sensory qualities of "Ngolo." The quality attributes evaluated were: visual appearance, texture, taste and flavour using hedonic scale of 1-9 where 4 is the limit of acceptability [14].

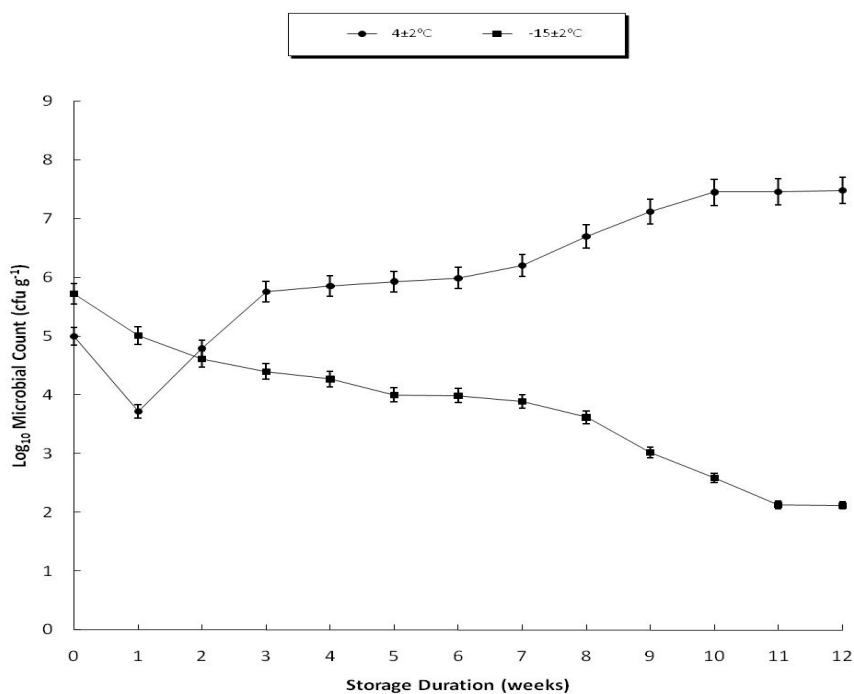
### 2.6 Statistical Analysis

Analysis of variance (ANOVA) of the obtained data was carried out to determine the significance of the mean differences at  $P = .05$  [15] based on software of SPSS version 15 for Windows (SPSS Inc. 2007).

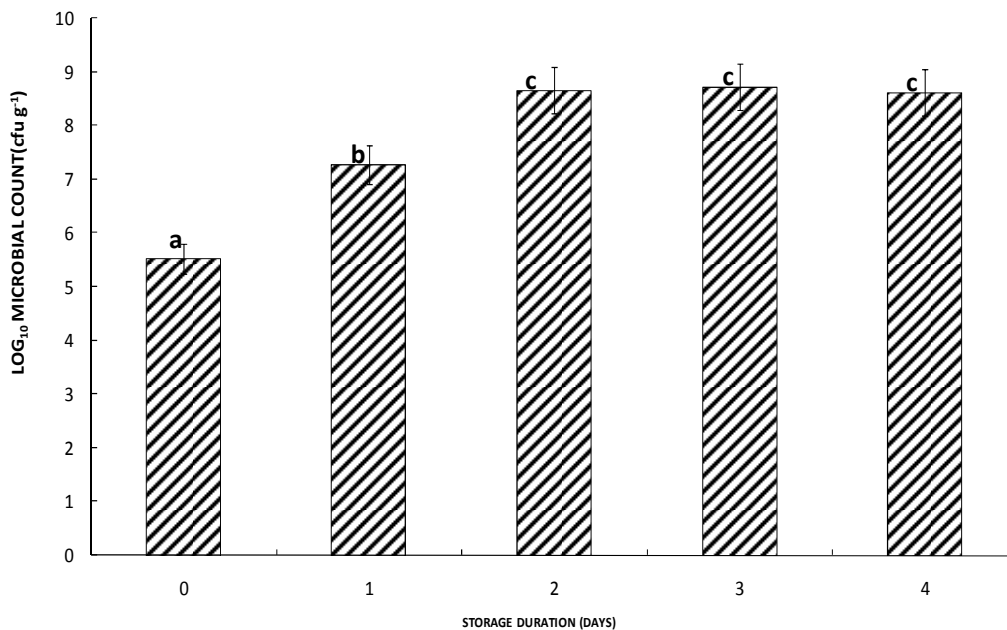
## 3. RESULTS

### 3.1 Microbiological Quality Changes in "Ngolo" Samples during Storage

A steady decrease in the total viable counts (TVCs) was observed in samples stored at -15±2°C but the decrease was more accentuated after the seventh week of storage (Fig.1). In contrast, the microbial dynamics of the "Ngolo" samples stored at 4±2°C showed a sharp decrease in the TVCs within the first week of refrigeration and thereafter, a gradual increase occurred attaining the maximum of 7.48 log CFU/g on the 12th week of storage (Fig. 1). On the other hand, the changes in total viable counts of the "Ngolo" samples stored at 29±2°C (short term evaluation) as shown in Fig. 2 where gradual increase led to the maximum load of 8.72 log CFU/g on day 3. "Ngolo" samples stored at -15±2°C showed the least bacterial diversity while samples stored at 29±2°C had the most heterogeneous bacterial genera (Table 1).



**Fig. 1. Changes in total viable counts of "Ngolo" samples stored under freezing (-15 ± 2°C) and refrigeration (4 ± 2°C) temperatures**  
 Each bar represents the mean ±SD of six determinations.



**Fig. 2. Changes in total viable counts of "Ngolo" samples stored at ambient temperature (29 ± 2°C).**  
 Each bar represents the mean ±SD of six determinations and bars carrying different letters indicate significant differences at P = .05

**Table 1. Microorganisms isolated from “Ngolo” samples during storage at different temperatures**

| Microorganisms            | -15±2°C | 4±2°C | 29±2°C |
|---------------------------|---------|-------|--------|
| <i>Bacillus</i> spp       | +       | +     | +      |
| <i>Pseudomonas</i> spp    | +       | +     | +      |
| <i>Flavobacterium</i> spp | -       | +     | +      |
| <i>Micrococcus</i> spp    | +       | +     | +      |
| <i>Staphylococcus</i> spp | -       | +     | +      |
| <i>Vibrio</i> spp         | -       | -     | +      |
| <i>Aeromonas</i> spp      | -       | +     | -      |
| <i>Escherichia coli</i>   | -       | -     | +      |

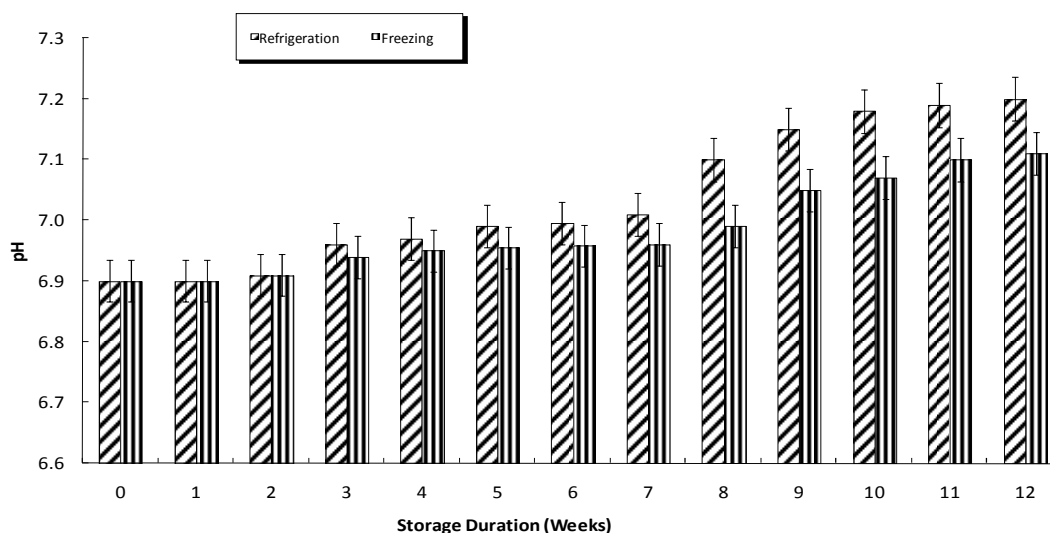
+ = Isolated, - = Not isolated

### 3.2 Physicochemical Quality Changes in “Ngolo” Samples during Storage

Little or no change occurred in the pH of samples stored at freezing and refrigeration temperatures, within the first two weeks (Fig. 3). But thereafter, the increase became more evident after the 7th week of freezing storage and 2<sup>nd</sup> week of refrigeration respectively, with the maximum of 7.11 and 7.23 occurring at the end of freezing and refrigeration storage (Fig. 3). The changes in pH of “Ngolo” samples stored at 29±2°C (short-term storage) as shown in Fig. 4 where a sharp decrease occurred on day 1 and thereafter, increased gradually to the maximum of 7.93 on day 4 of storage (Fig. 4).

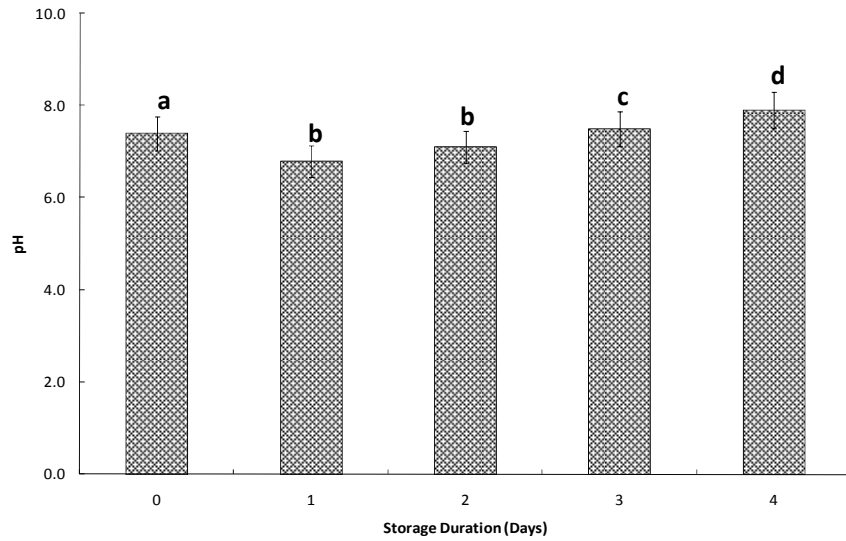
Changes in TVN and TMA values during storage at -15±2°C and 4±2°C are shown in Fig. 5 where

much lower values of TVN and TMA were obtained for samples stored at -15±2°C but increased gradually to 31 mgN/100 g and 25 mgN/100 g respectively on the 12<sup>th</sup> week of storage (Fig. 5). In contrast, changes in TVN and TMA values during storage at 4±2°C are shown in Fig. 5 where significant ( $P = .05$ ) increase from the initial TVN value of 12 mgN/100 g to 57 mgN/100 g occurred in the 12<sup>th</sup> week of storage while TMA increased from initial 9mgN/100 g to 34 mgN/100 g within the same period (Fig. 5). But samples stored at 29±2°C had low initial TVN and TMA values which increased steadily to the peak of 72mgN/100g and 48 mgN/100 g for TVN and TMA respectively on day 4 (short-term storage) (Fig. 6).



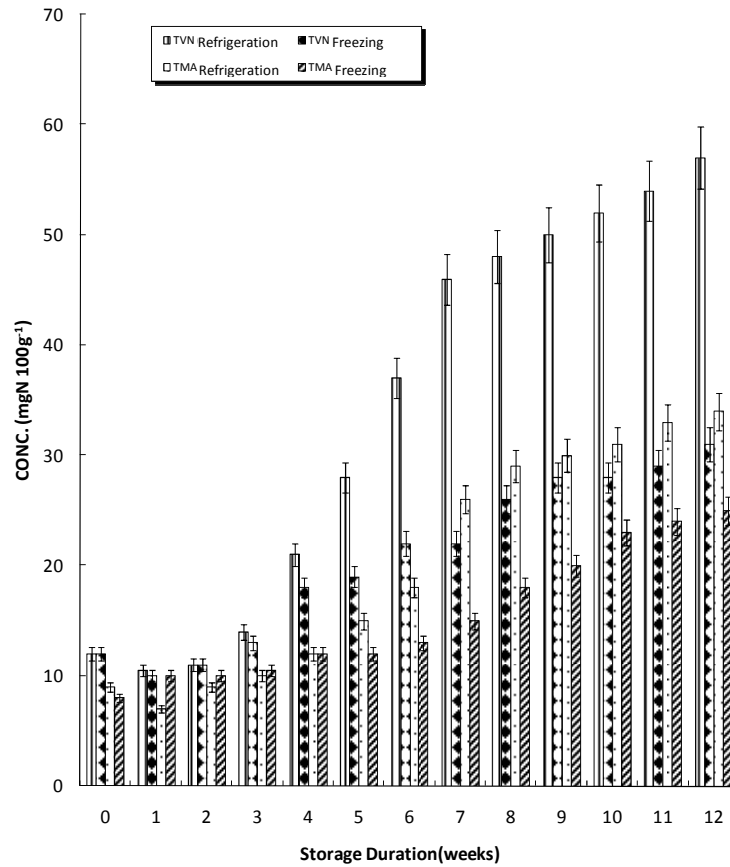
**Fig. 3. Changes in pH of “Ngolo” samples stored under freezing (-15 ± 2°C) and refrigeration (4 ± 2°C) temperatures**

Each bar represents the mean ±SD of six determinations



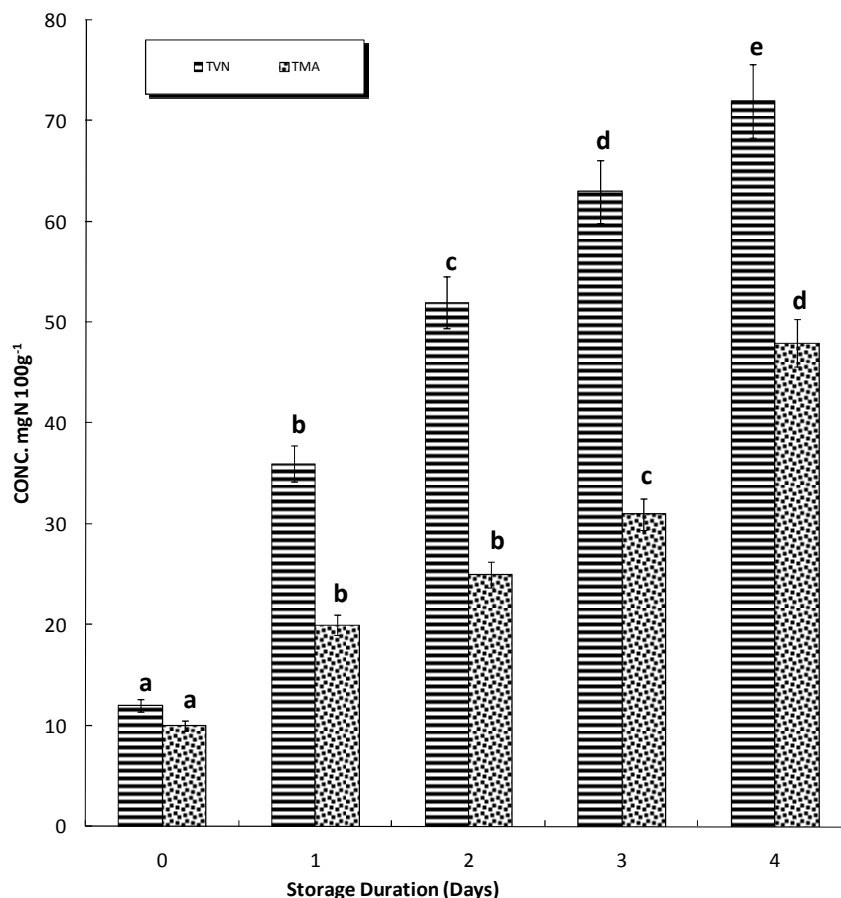
**Fig. 4. Changes in pH of "Ngolo" samples during storage at ambient temperature ( $29 \pm 2^\circ\text{C}$ )**

Each bar represents the mean  $\pm$ SD of six determinations and bars carrying different letters indicate significant differences at  $P = .05$



**Fig. 5. Changes in chemical indices (TVN and TMA) of "Ngolo" samples stored at under freezing ( $-15 \pm 2^\circ\text{C}$ ) and refrigeration ( $4 \pm 2^\circ\text{C}$ ) temperatures**

Each bar represents the mean  $\pm$ SD of six determinations



**Fig. 6. Changes in chemical indices (TVN and TMA) of "Ngolo" samples stored at ambient temperature (29±2°C)**

Each bar represents the mean ±SD of six determinations and bars carrying different letters indicate significant differences at  $P = .05$ .

### 3.3 Organoleptic Quality Changes in "Ngolo" Samples during Storage

The changes in sensory quality attributes of "Ngolo" samples during storage at  $-15\pm 2^\circ\text{C}$  are presented in Table 2 where initially, the sensory quality attributes of the freshly shucked samples were highly rated (acceptable) and remained highly acceptable throughout the duration of storage (Table 2). However, samples stored at  $29\pm 2^\circ\text{C}$  deteriorated significantly ( $P = .05$ ) and became unacceptable within 24h of storage (Table 3).

### 3.4 Correlations between Quality Indices of the Samples

Correlations between physico-chemical, microbiological indices and sensory scores of the "Ngolo" samples are shown in Table 4.

Significant positive correlation ( $r=0.9864$ ) occurred between TVN and storage duration as well as between pH and storage duration ( $r=0.9481$ ). Similarly, positive correlation of  $r=0.8145$  and  $r=0.7998$  were observed between total viable counts and storage duration as well as between total viable counts and total volatile nitrogen respectively. In contrast, the negative correlation of  $-0.9925$  and  $-0.8071$  were found between sensory scores (attributes) and TVN as well as between TVCs and sensory scores (Table 4).

## 4. DISCUSSION

Storage temperatures of foods are one of the most critical parameters for maintenance of quality attributes and microbial safety of foods [6,7,16]. The present findings have shown that storage of "Ngolo" samples at ambient

**Table 2. Changes in sensory quality of shucked “Ngolo” samples during storage at freezing (-15±2 °C) and refrigeration (4±2 °C) temperatures**

| Time (Weeks) | Temp/Organoleptic indices |           |           |           |            |           |           |           |
|--------------|---------------------------|-----------|-----------|-----------|------------|-----------|-----------|-----------|
|              | (-15±2 °C)                |           |           |           | (4±2 °C)   |           |           |           |
|              | Appearance                | Taste     | Flavour   | Texture   | Appearance | Taste     | Flavour   | Texture   |
| 0            | 7.3±0.03a                 | 7.6±0.03a | 6.9±0.09a | 7.2±0.04a | 7.3±0.12a  | 7.8±0.07a | 6.9±0.04a | 7.2±0.05a |
| 3            | 7.3±0.01a                 | 7.2±0.03a | 6.8±0.04a | 6.9±0.03a | 6.7±0.09a  | 5.8±0.12b | 5.9±0.06b | 6.1±0.07b |
| 6            | 7.1±0.04a                 | 7.0±0.06a | 6.4±0.06a | 6.7±0.03a | 4.3±0.04b  | 3.9±0.04c | 3.6±0.08c | 3.6±0.09c |
| 9            | 6.9±0.03a                 | 6.7±0.06b | 6.1±0.05b | 6.4±0.02b | 3.7±0.06b  | 3.3±0.06c | 3.4±0.05c | 3.2±0.03c |
| 12           | 6.7±0.09a                 | 6.5±0.05b | 5.8±0.07b | 6.1±0.05b | 3.0±0.08c  | 2.9±0.09d | 2.4±0.05d | 3.1±0.08c |

*Each value represents the mean ± SD of four determinations*

*Values having different letters in the respective columns are significantly different at P = .05*

**Table 3. Sensory attributes of shucked “Ngolo” samples stored at 29 ±2 °C for 48 hours**

| Storage time (HRS) | Temp/Organoleptic indices (29 ±2 °C) |            |           |           |
|--------------------|--------------------------------------|------------|-----------|-----------|
|                    | Appearance                           | Taste      | Flavour   | Texture   |
| 0                  | 7.3± 0.07a                           | 7.6±0.04a  | 7.3±0.03a | 7.2±0.05a |
| 12                 | 5.5± 0.06b                           | 5.8±0.07 b | 5.1±0.09b | 5.3±0.07b |
| 24                 | 3.7± 0.09c                           | 3.6±0.05 c | 3.4±0.04c | 3.7±0.04c |
| 36                 | 2.9± 0.04d                           | 2.5±0.08d  | 2.4±0.07d | 2.7±0.06d |
| 48                 | 2.0± 0.16d                           | 1.9±0.07e  | 1.9±0.40e | 2.2±0.07d |

*Each value represents the mean ± SD of four determinations. Values having different letters in the respective columns are significantly different at P = .05*

**Table 4. Relationship between storage duration and microbial, chemical and sensory indices of “Ngolo” samples stored at 29±2 °C**

| S/No | Variables correlated                               | Correlation values (r)* |
|------|--|-------------------------|
| 1.   | Total viable counts versus total volatile nitrogen | 0.79984                 |
| 2.   | Total viable counts versus storage duration        | 0.81450                 |
| 3.   | Total volatile nitrogen versus storage duration    | 0.98639                 |
| 4.   | pH versus storage duration                         | 0.94808                 |
| 5.   | Total viable counts versus sensory scores          | -0.80710                |
| 6.   | Total volatile nitrogen versus sensory scores      | -0.99247                |

*Each correlation value is based on overall mean of six determinations, \* Correlation coefficients (r) are significant at P = .05*



temperature resulted in more detrimental effects compared with either refrigeration or freezing storage (Figs. 1 and 2). The spoilage microorganisms encountered under ambient temperature storage ( $29\pm 2^\circ\text{C}$ ) were mostly mesophilic (Table 1) which indicate their ability to grow faster at this temperature being comparable to their optimum growth temperature. This partly explains the accelerated microbial increase observed in these samples accompanied by adverse physical and sensory changes such as softening of the tissues and putrid manifestations. Initially, the onset of spoilage included slight off-odour and loss of gloss of the "Ngolo" samples, resulting in strong off- odour coupled with dark colouration, sliminess and softness of the tissues by 24h under ambient temperature storage (Table 2). These deleterious changes could be attributed to microbial activities on proteinaceous, non-protein nitrogenous compounds and glycogen present in the "Ngolo" meat resulting in several metabolic products such as TVN and TMA [5,17].

The sharp decrease in microbial load within the first two weeks of refrigeration and freezing storage is probably attributed to induced long lag- phase periods of the microorganisms which may be partly ascribed to the elimination of mesophilic flora followed by the adaptation and growth of psychrotrophic [7,16]. However, the gradual increase in total viable counts during the refrigerated storage (Fig. 2) for all samples confirms that storage at  $4\pm 2^\circ\text{C}$  is not a preservative method for total inhibition of microorganisms [5,7,18]. In contrast, the microbial dynamics associated with the effects of freezing storage indicates a gradual and steady decrease in microbial population of the samples stored at  $-15\pm 2^\circ\text{C}$  (Fig. 2) which clearly indicates the impact of freezing with respect to microbial adaptation and lethal effects [16,19]. Thus, the non-detection of most microorganisms in samples stored at  $-15\pm 2^\circ\text{C}$  (Table 1) further demonstrates the variations in bacterial survival under different storage temperatures [5,19].

The initial decrease in the pH values of the samples stored at ambient temperature (Fig. 3) may be attributed to the breakdown of carbohydrate and glycogen in the "Ngolo" meat resulting in the formation of lactic acid. Such occurrences have earlier been observed in some seafoods [5,6,7]. However, the gradual increase in pH after 48h of storage may be attributed to

protein breakdown after depletion of carbohydrate and glycogen in the "Ngolo" meat which must have resulted in the production of amides, amines and ammonia as well as TMA and subsequently led to the pH increase with prolonged storage of "Ngolo" meat (Fig. 3). The more gradual increase in the pH of the samples stored at  $-15\pm 2^\circ\text{C}$ , as opposed to the accentuated increase after 7 weeks of refrigerated storage (Fig. 4) clearly, indicates the differential storage temperature effects on quality of seafoods. In addition, the rapid increase in TMA and TVN concentrations in samples stored at  $29\pm 2^\circ\text{C}$  (Fig. 5) is indicative of high proteolytic microbial activities (Fig. 1) and apparent spoilage based on TVN guidelines by Ward and Hackney [20]. Thus, the occurrence of significant positive correlations between TVCs and TVN as well as between pH and storage duration (Table 4) confirms the use of these quality indicators to predict the spoilage of 'Ngolo' and other seafoods as earlier reported [13,21].

Ward and Hackney [20] indicated that very good fresh shellfish should have a TVN value of less than 12 mgN/100 g while good quality shellfish has TVN value of 12-20 mgN/100 g. However, in general, acceptable/edible shellfish has TVN value of 20-25 mgN/100 g while decomposed and unacceptable shellfish has TVN value greater than 25 mgN/100 g. It is therefore, evident from these guidelines that "Ngolo" samples stored at  $29\pm 2^\circ\text{C}$  had become unacceptable within 24h of storage since the TVN value of 38 mgN/100 g was observed (Fig. 5). Shellfish contains Trimethylamine oxide (TMAO) in their flesh but during spoilage, the TMAO present in the fresh shellfish breaks down to TMA and ammonia [5] and 10-15 mgN TMA/100 g indicates the range of spoilage detection and unacceptability [5,6,13]. Thus, "Ngolo" samples stored at refrigerated and freezing temperatures had exceeded the limit of acceptability after the fifth and seventh weeks respectively. Furthermore, based on the European Council Directive 93/493 EEC [22] of critical value of TVCs 5 log cfu/g in cooked shellfish, the "Ngolo" samples stored at  $4\pm 2^\circ\text{C}$  and  $29\pm 2^\circ\text{C}$  were therefore considered microbiologically unsafe after two weeks and 24h respectively. Clearly, the changes in quality attributes (microbial, physicochemical and organoleptic) of "Ngolo" samples were significantly influenced by the different storage temperatures. These findings therefore underscore the benefits of adequate low temperature storage of "Ngolo" samples and the associated potentials for enhanced global trade.

## 5. CONCLUSIONS

The quality changes (microbiological, physicochemical and organoleptic) induced by the storage temperatures varied significantly with samples stored at  $-15\pm 2^{\circ}\text{C}$  remaining acceptable and safe throughout storage. But those subjected to storage at  $29\pm 2^{\circ}\text{C}$  were rejected and unsafe within 24h. Thus, the potential for enhanced international trade of "Ngolo" samples has been demonstrated.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

1. Deekae SN, Idoniboye-Obu TIE. Ecology and chemical composition of commercially important mollusks and crabs of the Niger Delta, Nigeria. *Environ Ecol*. 1995;13(1): 136-42.
2. Ovuru SS, Alfred-Ockiya JF. Occurrence of trace metals in *Penaeus notialis* from the Brass River systems of the Niger Delta. *Global J Pure Appl Sciences*. 2001;7(4):637-40.
3. Efiuvwevwere BJO, Ezeama CF. The bacteriological profiles of freshwater snail (*Pila ovata*) subjected to microcosms simulating local storage conditions. *World J Microbiol Biotechnol*. 2004;20:359-63.
4. Tan KS, Sigurdsson JB. A new species of *Thais* (*Gastropoda: Muricidae*) from Singapore and Peninsular Malaysia, *Raffles Bulletin of Zoology*. 1990;38(2):205-11.
5. Jay JM. *Modern Food Microbiology* 6<sup>th</sup> Edition. Chapman & Hall, New York. 2000;661.
6. Cao R, Xue C, Liu Q, Xue Y. Microbiological, chemical, and sensory assessment of pacific oysters (*C. gigas*) stored at different temperatures. *Czech J Food Sci*. 2009;27(2):102-8.
7. Mudoh MF, Parveen S, Schwarz J, Rippen T, Chaudhuri A. The effects of storage temperature in the growth of *Vibrio parahaemolyticus* and organoleptic properties in oysters. *Front Pub Health* Published 16 May; 2014.  
Available:<http://dx.doi.org/10.3389/fpubl.2014.00045>
8. Speck ML. *Compendium of methods for the microbiological examination of foods*. 3<sup>rd</sup> Edition. American Public Health Association, Washington DC. USA; 1992.
9. Martin RE, Carter EP, Flick Jr. GJ, Davis, LM. *Marine and freshwater products handbook* 963 pp. CRC Press, USA; 2000.
10. Harrigan WK, McCance ME. *Laboratory methods in food and dairy microbiology*, 2<sup>nd</sup> Edition. Academic Press Inc., London. 1976;322.
11. Sneath PHA, Mair NS, Sharpe ME. Holt JG. *Bergey's manual of systematic bacteriology*. Williams and Wilkins, Baltimore. USA 1986;2.
12. Pearson D. *The chemical analysis of food*. 9<sup>th</sup> Edition. Churchill Livingstone, London. 1992;464.
13. Sallam KI. Chemical, sensory and shelf life evaluation of sliced salmon treated with salts of organic acids. *Food Chem* 2007;101(2):592-600.
14. Meilgaard MC, Civille GC, Carr BT. *Sensory evaluation techniques*. 4<sup>th</sup> Edition. Springer Publishers, 280p 2004.
15. Snecdecor GW, Cochran WC. *Statistical methods*. The Iowa State College Press, Ames, Iowa. USA; 1976.
16. Ray B, Bhunia A. *Fundamental food microbiology*, 4<sup>th</sup> Edition. CRC Press, Boca Raton. USA; 2008.
17. Efiuvwevwere BJO, Amadi LO. Effects of preservatives on the bacteriological, chemical and sensory qualities of mangrove oyster (*Crassostrea gasar*). *British J Appl Sci Technol*. 2015;5(1):76-84.
18. Gomik SG, Albalat A, Macpherson H, Birkbeck H, Neil DM. The effect of temperature on the bacterial load and microbial composition in Norway lobster (*Nephrops norvegicus*) tail meat during storage. *J Appl Microbiol*. 2011;111(3): 582-92.
19. ICMSF (International Commission on Microbiological Specifications for Foods). *Microbial ecology of foods. Factors affecting life and death of microorganisms*, Academic Press, New York, USA. 1980;1.
20. Ward DR, Hackney C. *Microbiology of Marine food products*. Van Nostrand Reinhold Publishers, New York. 1991; 449.

21. Zeng QZ, Thorarinsdottir KA, Olafsdottir G. Quality changes of shrimp (*Pandalus borealis*) stored under different cooling conditions. J Food Sci. 2005;70(7):s459-s66.
22. FAO, 1993. Corporate Document Repository: Assessment and management of seafood safety and quality. (EEC 93/493).

© 2016 Oruwari and Efiuvwevwere; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

*Peer-review history:*  
*The peer review history for this paper can be accessed here:*  
<http://sciencedomain.org/review-history/15666>