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Full Length Research Paper

Water activity, microbial and sensory evaluation of smoked fish (*Mormyrus caschive* and *Oreochromis niloticus*) stored at Ambient Temperature, Terekeka-South Sudan

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The effect of smoking kilns on water activity, microbial load and sensory attributes was evaluated to adopt suitable technology that maintains the quality of fish in South Sudan. A total of 300 fresh *Mormyrus caschive* and *Oreochromis niloticus* were purchased, 36 fresh samples were iced and the remaining 264 fish samples were divided into two batches for pit and *chorkor* smoking. The experiment was done twice in a completely randomized design. Fish samples were analyzed for water activity using Lab-Swift Meter, microbial load using standard microbiological methods and sensory characteristics using 9-point hedonic grading scale. Result revealed that, water activity in pit smoked fish increased at a rate of 1.7 times faster than in *chorkor* smoked samples during storage. Corresponding to water activity, plate count equally increased at a rate of 1.7 times faster for pit smoked *M. caschive* and 1.1 times for *O. niloticus* than samples smoked using *chorkor*. Similarly, mould load increased at a rate of 1.5 times faster for pit smoked *M. caschive* and 2.2 times for *O. niloticus* than samples smoked using *chorkor* oven. Overall, *chorkor* smoked fish had significantly better sensory parameters than pit smoked samples after one month's storage. Therefore, *chorkor* kiln produced quality smoked fish in terms of microbial load and organoleptic parameters, and the study recommends its adoption for artisan fisheries in South Sudan. However, microbial characteristics of smoked fish need to be further examined for a period exceeding a month to determine microbiological quality and establish fish self-life in order to maintain the quality and safety of fish and fisheries products.

Key words: Water activity, microbial load, sensory attributes, smoking technologies, shelf life.

INTRODUCTION

Fish is an important source of food and income to a large section of population in the developing world (FAO, 2015). In Africa, about 35 million people, depend wholly or partly on the fishing sector, mostly the small-scale

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fisheries, for their livelihoods (Adeyeye and Oyewole, 2016). In particular, fish is an important and cheap source of protein to low income earners in developing economies (Nunoo and Kombat, 2013). Fish forms more than 50% of the essential animal protein and mineral intake for over 400 million people from developing countries (FAO, 2016). In South Sudan, over 1.7 million people depend on fisheries for their livelihoods (FAO, 2018). The immense contribution and importance of artisan fishing to livelihoods in South Sudan increased from 6.8 to 10.2% from 2015 to 2016, indicating greater reliance on this source of income (FAO, 2018). In terms of employment and income source, FAO (2016) estimated that over ten billion people in the developing world are engaged in fisheries activities mainly as fishers, processors and vendors. However, fisherfolks mostly processors are faced with high post-harvest losses due to lack of proper processing and preservation techniques (Adeyeye et al., 2016; Amegovu et al., 2017). Poor handling of fish catches leads to significant physical, quality, nutritional and economic losses (Adeolu et al., 2017).

Equally, fish post-harvest handling provides livelihoods to millions of people and contributes significantly to foreign exchange of many countries in the world (FAO, 2016). Fish processing and preservation are important due to the fact that fish is highly susceptible to deterioration after harvest (Olopade et al., 2013; Getu and Misganaw, 2015), and to prevent quality, nutrient and economic losses (Golub, 2014). Hygienic handling of fish is essential to attain the best quality and highest possible profits (Muhame et al., 2020). Despite the fish vast importance, fishing industry suffers from enormous post-harvest losses which are estimated at 30% in dry season and 40% in rainy season (FAO, 2016). Fish post-harvest losses have a profound adverse impact on rural fishing population whose livelihoods often depend on fisheries activities (Adeyeye and Oyewole, 2016).

Lack of proper infrastructure for fish post-harvest handling is a major challenge facing rural fishing communities of South Sudan (Amegovu et al., 2017). As such, improved processing and preservation technologies are sought to reduce adverse effects of fish post-harvest losses. Among the traditional methods of salting, sun drying and fermentation, fish smoking is the main type of fish preservation technique used in Terekeka (UNIDO, 2015). This is due to the fact that most consumers prefer smoked fish to sun dried, wet salted and fermented fish in addition to technical simplicity (Magawata and Musa, 2015). Fish smoking is also commonly practiced in remote areas where delivery of fresh catches to distant markets is not permitted due to poor roads and lack of cool chain transport facilities (Adeyeye and Oyewole, 2016). The need to improve smoking technologies in such areas is of a paramount importance to reduce fish post-harvest losses (Emere and Dibal, 2013; Adeolu et al.,2017). Effective and efficient fish smoking indeed, maintain the quality and safety of fish due to antimicrobial, antioxidants and preservatives contained in the smoke (Omodara et al., 2016; Pemberton-Pigott et al., 2016; Yusuf and Hamid, 2017). The safety and quality of smoked fish help consumers in making decisions on processed fish products (Nunoo and Kombat, 2013), which also influence the market prices (Nguvava, 2013).

Fish smoking and drying reduce moisture, concentrates nutrients (Kumolu-Johnson et al., 2010), and reduce water activity in processed fish products (Pal et al., 2016). Well smoked fish with low water activity of less than 0.50 and moisture content between 15 and 25% is noted to inhibit growth of pathogenic microorganisms in the products (Francisca et al., 2010). During smoking process where salt is used, micro-organisms at the surface of fish are potentially inhibited (Oparaku and Mgbenka, 2012). Nyarko et al. (2011) reported mean microbial load in salted-smoked fish in the range of 4.79 to 6.52 log cfu/g which are within the acceptable limits for microorganisms in preserved foods as recommended by the International Standards. Olayemi et al. (2012) reported total bacterial and mould counts of 2.0×10^4 and 0.7×10^4 cfu/g, respectively, depending on fish species. Olayemi et al. (2012) added that microbial load is associated with the location of fish harvest, method of fishing, amount of salt used, smoking conditions, smoke chemicals and handling practices. Yusuf and Hamid (2017) noted that, contamination of dried fish by microorganisms occurs during processing, storage, transportation and during sale at open air markets.

Proper handling of fish during processing is noted to lower microbial count in the final products (Huong, 2014). However, the effect of smoking technologies on water activity, microbial load and sensory characteristics of smoked fish during storage at ambient temperature has not been determined in South Sudan. This study was therefore, conducted to assess water activity, microbial load and sensory attributes of pit and *chorkor* smoked *Mormyrus caschive* (L) and *Oreochromis niloticus* (L) stored at ambient temperature to enable fisherfolks to adopt suitable smoking technology that maintains quality and safety of smoked fish in South Sudan.

MATERIALS AND METHODS

The study area

The study was conducted in June, 2018 in Terekeka, South Sudan. Terekeka State is located at 70 km north of Juba on the western bank of the Nile (Miller and Benansio, 2011). Terekeka lies within Latitudes 5°26'38"N and Longitudes of 31°45'02"E, respectively (FAO and WFP, 2019). The state covers an area of about 10,358.95 km^2 and is occupied by an estimated population of more than 246,483 (South Sudan Centre for Census, Statistics and Evaluation, 2018). Terekeka has tropical climate with comparatively small seasonal variation of temperature, humidity and wind throughout the year. Terekeka usually receives rainfall between the months of April to November with an average annual rainfall of 907 mm. The area experiences dry periods between the months of December and March with an average annual temperature of

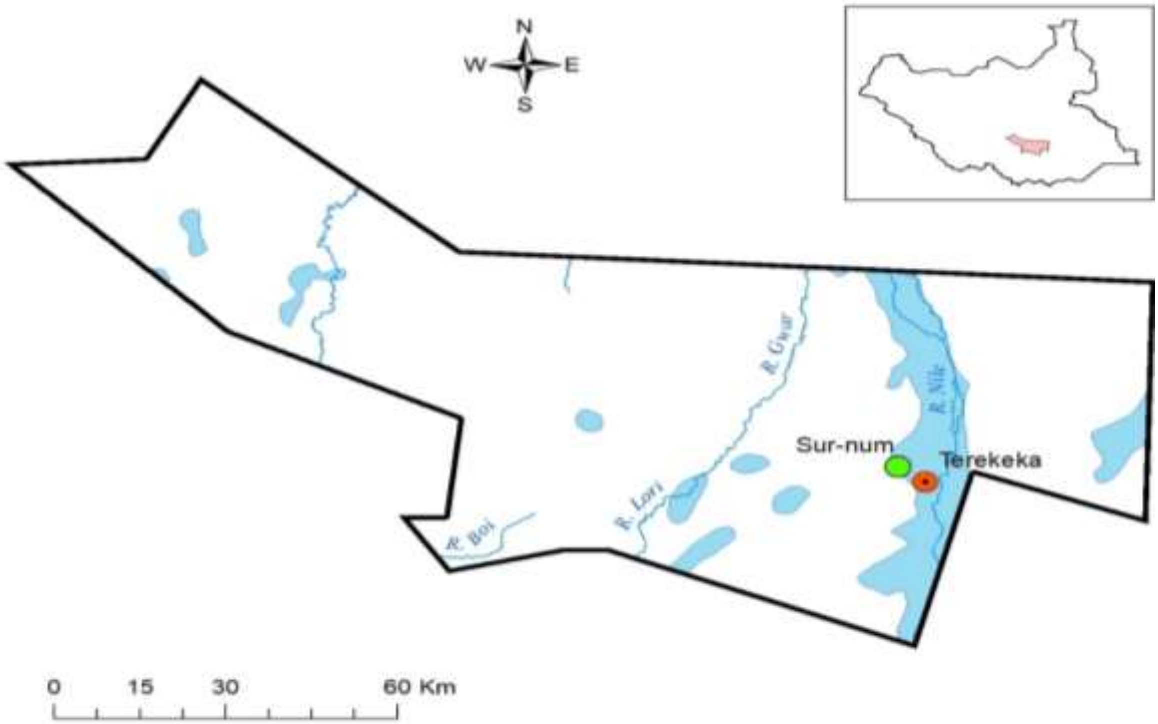


Figure 1. Map of the study area (Sur-num Landing Site, Terekeka State-South Sudan in Africa).

Table 1. Systematic positions, common and local names of the two fish species used for the study.

Number	Family	Genus	Species	Common name
1	Mormyridae	Mormyrus	*Mormyrus caschive*	Elephant snout
2	Cichlidae	Oreochromis	*Oreochromis niloticus*	Nile Tilapia

27.7°C. It is within the dry season that most people are actively involved in fisheries activities. However, plentiful fish catch occurs during the months of June to August particularly when the flood recedes (Figure 1).

Study design

The study was conducted in two phases, field experiment and laboratory analysis. Physical parameters of the two fish species were recorded in the field. Systematic positions according to Linnaeus (1758) and common names of the two fish species used for this study are shown in Table 1.

The lengths and weights were measured using meter rule and digital weighing scale, and reported as Means ± Standard Deviation. Experimental smoking using pit and *chorkor* smoking kilns was conducted twice in a completely randomized design. Women fish group *chorkor* oven was used to represent improved smoking technology. The *chorkor* measuring 2 m long, 1 m wide and 1 m high with 3 trays were attached after every 30 cm from base to top. The *chorkor* was constructed using unbaked bricks and the interior part plastered with clay soil. The top part roofed with perforated flat iron sheets. It has two inlets at the base for aeration and smoke production by burning firewood. The smoking chamber of *chorkor* oven has a movable door that remained closed except during monitoring.

Traditional pit was constructed alongside improved *chorkor* oven. Its dimensions were 1 m long, 0.5 m wide and 0.5 m high as practiced by fisherfolks in the area. Four wooden planks were placed at the edges where a wire mesh sits. During fish smoking, flat iron sheet was used to cover the samples. Fish smoked samples from the two kilns were wrapped in aluminium foils, labeled for easy identification, packed in containers and transported by vehicle to the laboratory in Makerere University, Uganda for analyses. While in the laboratory, samples were removed from the aluminium foil and kept at ambient temperature during the analysis to depict storage conditions provided by fish vendors in South Sudan. In the laboratory, three study parameters were determined to compare the two smoking technologies: water activity, microbial load and sensory characteristics (Figure 2).

Sampling, processing and analysis procedures

To cater for all the study parameters, a total of 300 fresh *M. caschive* and *O. niloticus* were purchased from fishermen at Sur-

Figure 2. The structures of Pit (left) and *Chorkor* (right) smoking ovens.
Source: Photograph taken by Charles Mondo on 26, June 2018.

num landing site situated at about 1 km from Terekeka town. Fish processing was done immediately after delivery. Fresh fish samples were kept in cooler boxes packed with ice blocks instantly after being harvested at the fishing grounds. Purposeful fish smoking using improved *chorkor* and traditional pit was conducted at Terekeka landing site. From the procured samples, 18 specimens of each species were ice stored at 4°C and transported to the laboratory where 6 specimens from each species were randomly picked and destined for microbial analysis. The remaining 282 specimens were divided equally into two batches, taking into account the species for pit and *chorkor* experimental smoking using *Acacia seyal,* the dominant tree species for smoking fish in the area.

All fish were washed to remove slime, descaled, eviscerated and rewashed thoroughly with clean water to remove blood. Prepared fish samples were immersed in freshly prepared salt solution (a mixture of 100 g salt in 10 L of clean water) for 15 min followed by draining for 15 min. Fire was set in pit and *chorkor* kilns to generate smoke heat by burning *A. seyal* wood. The pre-treated fish samples were randomly loaded on the wooden trays and wire mesh on top of *chorkor* and pit, respectively. The desired temperature of 60 to 80°C was maintained manually by a thermometer until the fish were smoke dried. During smoking, the position of fish samples in the wooden trays were changed in *chorkor* to attain uniformity of the products and turned upside down in pit kiln in mid periods in order to make samples smooth and steady in texture and appearance. Smoked fish were cooled for 12 h at ambient temperature. Smoked samples were wrapped in aluminium foils, labeled for easy identification, packed in containers and transported by road transport to the laboratory in Makerere University, Uganda for analyses. While in the laboratory, smoked fish samples were removed from the aluminium foil and kept at ambient temperature during the analysis period to depict the storage conditions provided by fish vendors in South Sudan.

Determination of water activity in smoked fish samples

For water activity, 10 g of fish muscles were aseptically removed from each fish species. The portion was placed in a round internal plate and inserted into the water activity meter (Lab Swift Bench Model Water Activity Meter, BS ISO 21807:2004; Public Health England, 2017). The meter set at 20°C, automatically read the water activity value in the sample after 5 min time.

Determination of microbial load in fish samples

Standard methods were used for identification and enumeration of microorganisms (ISO, 2003; 2008; ICMSF, 1986; BAM, 2005). Iron Agar and Dicloran Rose Bengal Chloramphenicol Agar used in this study were obtained from Oxoid Limited, England. These media were prepared and sterilized according to the manufacturer's instructions. Sterility control plates of each medium and diluent were made by incubating them overnight at a suitable temperature.

Sample preparation for microbial load determination

From the fish samples, a 10 g of each sample was removed from the muscle and aseptically weighed on an electronic balance. The weighed samples were blended in a stomacher 400 (Lab. Blender, London, UK) and transferred individually into 180 mL dilution bottles (Borosilicate-resistant glass with rubber stopper) containing 90 mL of sterile peptone water (0.1%) for suspension. This was done for samples of the two fish species obtained from each smoking technology and was taken as the original stock. The content was homogenized using vortex mixer and a dilution of 1:10 (10^{-1}) and 1:100 (10^{-2}) were obtained using standard serial dilution method (David and Davidson, 2014). The spread plate technique was used to culture microorganisms (Yusuf and Hamid, 2017). After incubation, the number of colonies on a dilution plate between 30 and 300 colonies were counted and the number of microorganisms was determined using ISO 4833:2003 (ISO, 2003) formula:

$$N = \frac{\sum C}{V(n1 + 0.1n2)d}$$

where *N*= number of colonies per mL or gram of the sample, $\sum C$= sum of all colonies counted on the plates containing 30-300 colonies, *n1*= number of plates counted in the lower dilution, *n2*= number of plates counted in the higher dilution, *d*= value corresponding to the dilution from which the first counts were obtained and *V*= volume of inoculums used. The total viable count and mould count were recorded as colonies formed by microorganisms in the inoculated plates. The means of duplicate microbial count in colony forming units were determined and transformed to $\log_{10}$ cfu/g for statistical analyses. Media and air control plates were also prepared and carried out parallel to the analysis as internal quality control.

Table 2. Initial mean values of water activity and microbial load recorded in smoked *M. caschive* and *O. niloticus*.

Parameter	*Mormyrus caschive*		*Oreochromis niloticus*	
Smoking types	**Pit**	***Chorkor***	**Pit**	***Chorkor***
Water activity (a_w)	0.62	0.53	0.62	0.56
Total plate count (log_{10} cfu/g)	2.12	1.51	2.50	1.54
Mould count (log_{10} cfu/g)	1.65	1.00	1.65	1.30

Values are means of duplicate determination of homogenized fish muscles.

Enumeration and identification of microorganisms

For total aerobic plate count, 1 mL of prepared dilution from each sample was spread on plates inoculated with Standard Iron Agar (IA) in duplicates. Plates were then aerobically incubated in an inverted position at 35 ± 2°C. Colonies were counted after 48 h of incubation using Reichert Dark Field Quebec Colony Counter. For moulds' isolation, a standard method of ISO 21527-1:2008 was used with minor modifications. Briefly, 1 mL of each dilution was spread on the surface of plate inoculated with Dicloran Rose Bengal Chloramphenicol Agar (DRBC Agar; Category 1160) using a sterile glass rod. DRBC Agar inoculated plates were incubated uprightly at 25 ± 1°C. Counting of mould colonies was done after 5 days of incubation using Quebec Colony Counter. Mould colonies were recorded as colony forming units per gram (cfu/g) of fish samples. The means of duplicate microbial count in colony forming units were calculated and transformed to log_{10} cfu/g for statistical analyses.

Evaluation of organoleptic attributes of smoked fish

The organoleptic attributes: appearance, aroma, flavour, taste, texture, and the overall acceptability of smoked fish were evaluated. Preference and paired comparison test as recommended by FAO (1999) in the Codex guidelines for sensory evaluation of fish and shell fish in laboratories (CXG 31-1999) were used. The sensory attributes were assessed by a control test panel of 20 individuals using 9-point hedonic grading scale as described by Aba and Ifannyi (2013). Grading scales were described as "poor" for attributes assigned value (1 to 3), "fairly good" (4-5), "good" (6-7) and "very good" (8-9). Twenty staff members (control test panel) consisting of 10 males and 10 females of age range 25 to 45 years old from the National Agriculture Research Laboratories in Kawanda constituted the sensory evaluation panel. As a precaution, panelists were given 10 min break interval to wash their mouths using distilled water after every sample assessment to avoid carry over effect. Panelists were also prevented from communicating to each other during the assessment by allotting a booth to each member.

Preparation of smoked fish samples for sensory evaluation

Pit and *chorkor* smoked fish samples were differently cooked in a microwave container for 10 min at 121°C and allowed to cool to a comfortable tasting temperature. Then coded with 3 figures for example: 101, 102, 103 and 104 for pit and *chorkor* smoked *M. caschive* and *O. niloticus* assessed on day 0 (week one). Coded samples were randomly placed in a tray and a glass of clean water provided for panelists to cleanse their palate after each sample taste. Prepared fish samples were served to each booth where a participating panelist was assigned. Panelists indicated scores for preference and compare between samples obtained from pit and *chorkor* technologies. Assessors noted organoleptic tests and general acceptability of the four specimens and recorded their observation by assigning a value to each specimen.

Statistical analyses

Data collected from the study was analyzed using R statistical package (R Core Team, 2018). A two-way analysis of variance (ANOVA) was used to test the difference in water activity, microbial counts and sensory attributes recorded in fish samples smoked using *chorkor* and pit to determine the effect of smoking technologies on the quality parameters of fish. Tukey's Honest Significant Difference test was performed where the means of the two groups under comparison were significantly different. Pearson Correlation Analysis (PCA) was done to establish the relationship between the factors: water activity and microbial growth associated with sensory attributes of smoked fish. Statistical significance level was studied at $P \leq 0.05$.

RESULTS

Physical parameters of the two fish species

Average lengths and weights of the two fish species were 28.17±1.17 cm, 133.06±3.60 g and 23.31±1.01 cm, 126.73±1.86 g for *M. caschive* and *O. niloticus,* respectively.

Water activity and microbial load in smoked *M. caschive* and *O. niloticus*

The initial mean water activity values in pit smoked fish were higher than values recorded in *chorkor* smoked *M. caschive* and *O. niloticus,* respectively (Table 2). Equally, the initial total plate count was significantly higher in pit than in *chorkor* smoked *M. caschive* and *O. niloticus,* respectively. Similarly, the initial mould count was significantly higher in pit than in *chorkor* smoked *M. caschive* and *O. niloticus,* respectively. Generally, *M. caschive* and *O. niloticus* smoked using *chorkor* oven had significantly lower microbial load than pit smoked samples ($P<0.05$).

Water activity (a_w) determined in smoked fish during storage

Generally, water activity increased with increase in

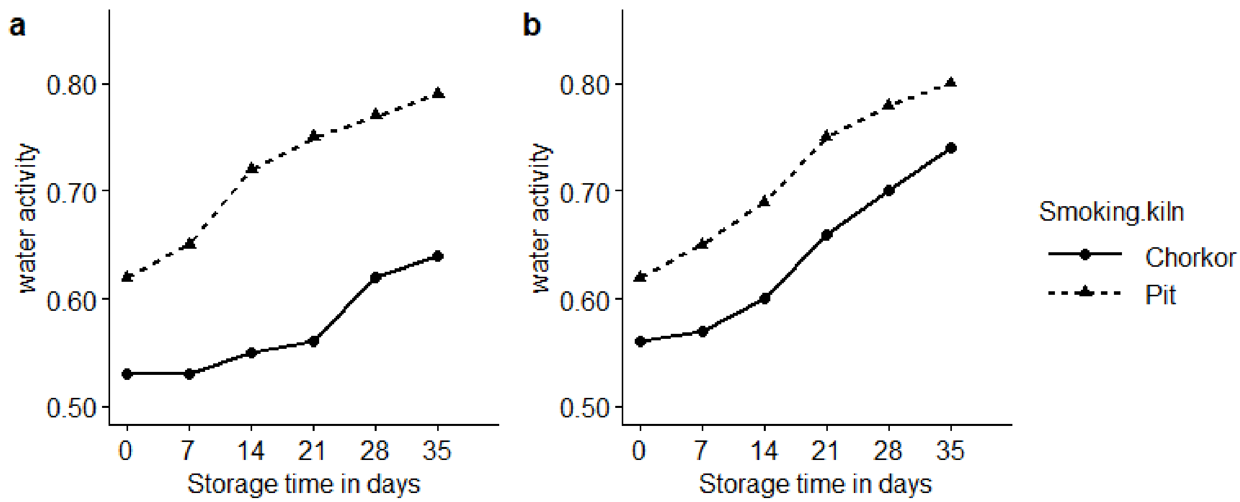


Figure 3. Trend in water activity recorded in pit and *chorkor* smoked *M. caschive* (a) and *O. niloticus* (b) stored for 35 days at room temperature.

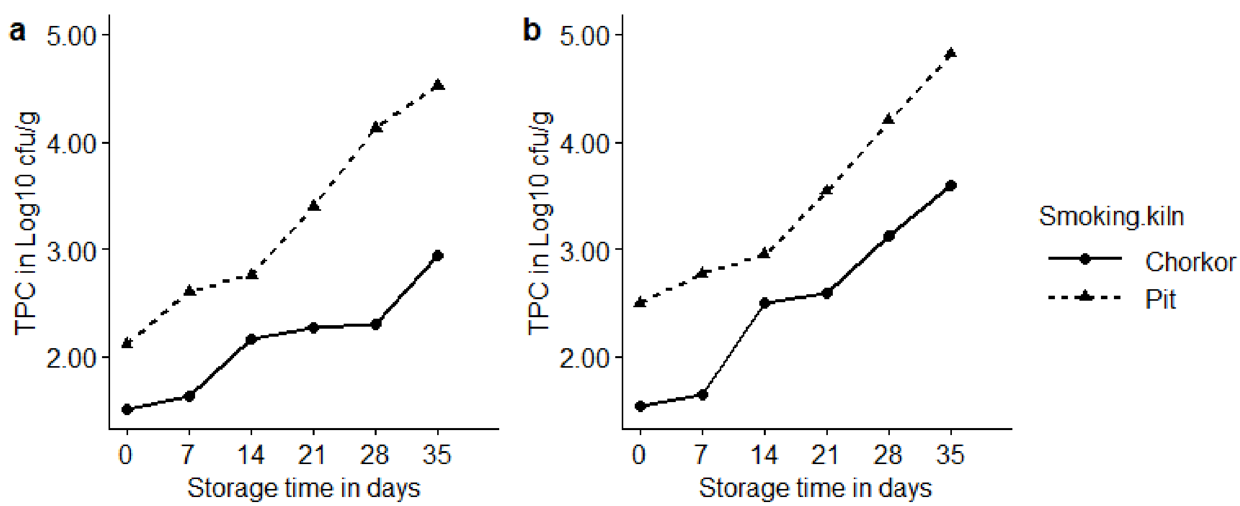


Figure 4. Trend in total plate count recorded in pit and *chorkor* smoked *M. caschive* (a) and *O. niloticus* (b) stored for 35 days at room temperature.

storage over the 35 days keeping time (Figure 3). The average rate of increase in water activity in pit smoked fish (0.005/week) was significantly higher than in *chorkor* smoked samples (0.003/week, P<0.05). With regards to smoking technologies, mean water activity recorded in pit smoked fish was significantly higher than that measured from *chorkor* specimens (P<0.05, Figure 3a and b).

Total plate count of smoked fish during storage at ambient temperature

The study recorded an increasing trend in microbial load, total plate count (TPC) in the muscles of smoked fish during storage at room temperature (Figure 4a and b). Total plate count increased significantly with increase in storage time. With regards to the technologies, pit smoked *M. caschive* has significantly higher total plate count than *chorkor* smoked samples in the entire storage period (P<0.05). The growth rate of microorganisms in smoked fish products during storage revealed that, pit smoked *M. caschive* has higher average growth rate of total viable count (0.069 log_{10} cfu/g/week) than *chorkor* smoked fish samples (0.041 log_{10} cfu/g/week, P<0.05). Similarly, the total plate count of smoked *O. niloticus* increased with increase in storage time (Figure 4b). Pit

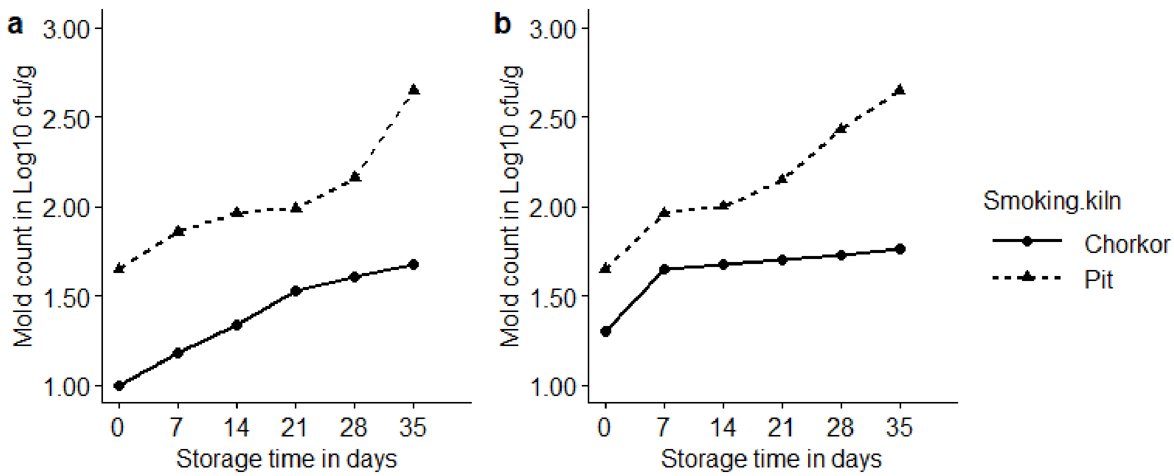


Figure 5. Trend in mould count recorded in pit and *chorkor* smoked *M. caschive* (a) and *O. niloticus* (b) stored for 35 days at room temperature.

smoked *O. niloticus* also has significantly higher total plate count than *chorkor* smoked samples (P<0.05). With regards to growth rate, pit smoked *O. niloticus* has higher average growth rate (0.066 log_{10} cfu/g/week) than *chorkor* smoked samples (0.059 log_{10} cfu/g/week). The increase in microbial load in smoked fish during storage strongly correlated (r^2=0.94) with increase in water activity. With regards to storage, the fourth and fifth weeks of storage showed higher water activity and microbial load than the first three weeks.

Mould count of smoked fish during storage at ambient temperature

The study observed an increasing trend in mould count in the muscles of smoked fish during storage at room temperature (Figure 5). Moulds increased significantly with increase in keeping time (P<0.05). With regards to the technologies, pit smoked *M. caschive* has significantly higher mould count than *chorkor* smoked samples during the entire storage period (P<0.05). The average growth rate of mould was higher in pit smoked *M. caschive* (0.029 log_{10} cfu/g/week) than in *chorkor* smoked samples (0.019 log_{10} cfu/g/week). Similarly, smoked *O. niloticus* has significantly higher mould counts than *chorkor* smoked fish samples over storage time. The average growth rate of moulds was higher in pit smoked *O. niloticus* (0.029 log_{10} cfu/g/week) than in *chorkor* smoked samples (0.013 log_{10} cfu/g/week).

Sensory evaluation of smoked *M. caschive* and *O. niloticus*

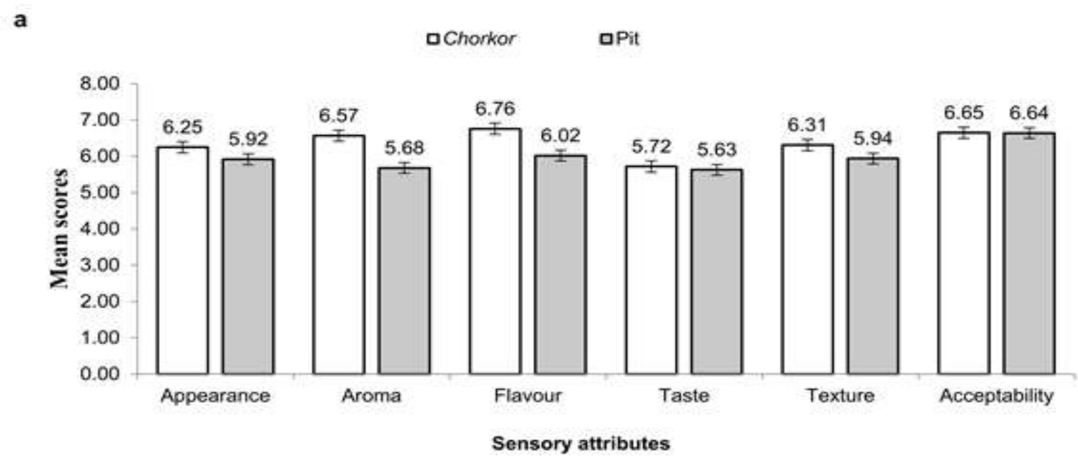
The mean score of organoleptic sensory tests of *chorkor* smoked samples were significantly higher than pit smoked *M. caschive* (P<0.05; Figure 6a). Meanwhile, the mean score for overall acceptability of fish smoked using both technologies did not significantly differ. With regards to *O. niloticus,* the mean scores of all the organoleptic sensory attributes of *chorkor* smoked fish were significantly higher than specimens smoked using pit kiln (P<0.05; Figure 6b). Comparison of means using Tukey's test showed a significant difference in the means of pit and *chorkor* smoked fish (P<0.05). All the organoleptic attributes have mean scores above 5, the critical limit for acceptability of smoked fish products under quality assessment.

With regards to the attributes, a decreasing trend was noted in the colour or appearance mean scores of smoked fish during storage (Table 3). The dark-brown colour of smoked *M. caschive* and *O. niloticus* decreased with increase in storage time. In terms of technologies, *chorkor* smoked *M. caschive* and *O. niloticus* had higher colour mean scores than pit smoked fish products. The difference was significant on the first week of the storage (P<0.05).

There was a decreasing trend in aroma of smoked fish with increase in storage time (Table 4). On the third and fourth week, the unique aroma of smoked fish decreased significantly with increase in storage time with *chorkor* smoked *M. caschive* and *O. niloticus* having better aroma than pit smoked fish (P<0.05).

The smoky flavour of pit and *chorkor* smoked *M. caschive* and *O. niloticus* decreased with increase in keeping time (Table 5). With regards to the technologies, *chorkor* smoked *M. caschive* and *O. niloticus* had better flavour than pit smoked fish products.

Similarly, the pleasant taste in smoked fish decreased with increase in storage time. Concerning the technologies, *chorkor* smoked fish had better mean taste



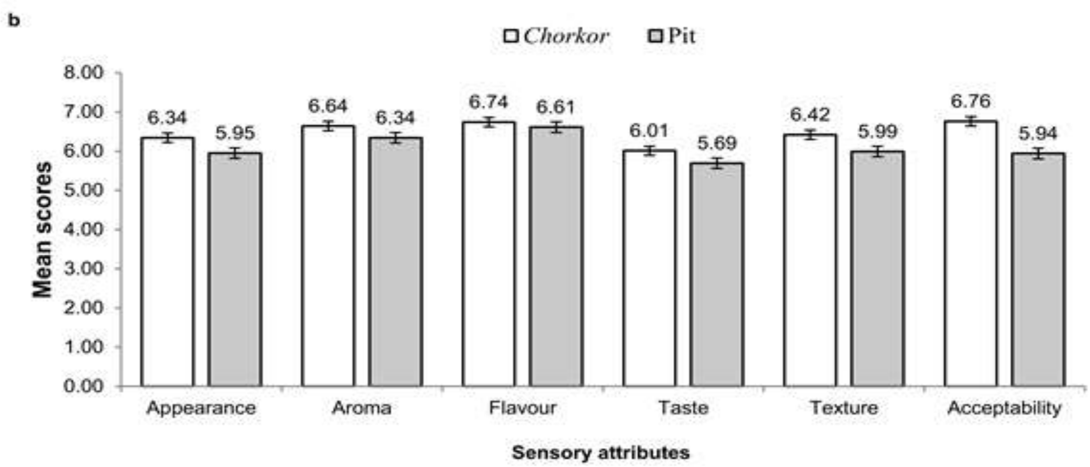


Figure 6. Overall sensory attributes of pit and *chorkor* smoked *M. caschive* (a) and *O. niloticus* (b) stored for 28 days at room temperature

Table 3. Trend in appearance or colour mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at room temperature.

Parameter	Storage time (Days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Appearance	0	6.20 ± 1.24^{b}	6.50 ± 1.67^{ab}	6.60 ± 1.98^{a}	7.00 ± 1.34^{a}
	7	5.80 ± 2.24^{b}	5.80 ± 2.04^{b}	6.10 ± 2.25^{b}	6.25 ± 1.41^{b}
	14	5.75 ± 1.80^{b}	5.75 ± 2.05^{b}	5.65 ± 1.90^{b}	5.95 ± 1.28^{b}
	21	5.30 ± 1.45^{c}	5.35 ± 1.18^{c}	5.10 ± 2.17^{c}	5.65 ± 1.23^{b}
	28	5.10 ± 2.17^{c}	5.20 ± 1.79^{c}	5.00 ± 1.75^{c}	5.20 ± 2.55^{c}

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

scores than pit smoked fish products (Table 6).

There were textural changes in pit and *chorkor* smoked fish. The mean textural scores in fish smoked using pit and *chorkor* technologies decreased with increase in storage time, and *chorkor* smoked fish had higher mean textural scores than pit smoked fish products (Table 7).

Table 4. Trend in aroma mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at room temperature.

Parameter	Storage time (Days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Aroma	0	6.80 ± 1.74^{a}	6.80 ± 1.77^{a}	6.80 ± 1.77^{a}	7.20 ± 1.61^{a}
	7	6.05 ± 1.93^{b}	6.70 ± 1.87^{a}	6.70 ± 1.87^{a}	6.75 ± 1.45^{a}
	14	5.85 ± 1.90^{b}	6.65 ± 1.27^{ab}	6.65 ± 1.27^{ab}	6.70 ± 2.07^{a}
	21	5.25 ± 1.45^{c}	6.50 ± 1.54^{ab}	6.50 ± 1.54^{ab}	6.55 ± 1.15^{ab}
	28	5.00 ± 1.35^{c}	6.20 ± 1.82^{b}	6.20 ± 1.82^{b}	6.00 ± 1.38^{b}

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

Table 5. Trend in flavour mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at ambient temperature.

Parameter	Storage time (Days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Flavour	0	7.25 ± 1.33^{b}	7.50 ± 1.43^{a}	7.10 ± 1.37^{b}	7.25 ± 1.37^{b}
	7	6.55 ± 1.82^{bc}	6.90 ± 1.59^{b}	6.80 ± 1.51^{b}	6.90 ± 0.85^{b}
	14	6.10 ± 1.33^{c}	6.65 ± 1.35^{b}	6.50 ± 1.79^{b}	6.65 ± 1.27^{bc}
	21	5.15 ± 1.60^{d}	6.45 ± 1.61^{c}	6.40 ± 1.70^{c}	6.50 ± 1.05^{bc}
	28	5.05 ± 1.54^{d}	6.30 ± 1.81^{c}	6.25 ± 1.80^{c}	6.40 ± 1.27^{c}

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

Table 6. Trend in taste mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at ambient temperature.

Parameter	Storage time (Days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Taste	0	6.45 ± 2.11^{b}	6.90 ± 1.29^{a}	7.05 ± 2.01^{a}	7.25 ± 1.59^{a}
	7	6.25 ± 1.59^{b}	6.70 ± 1.81^{a}	6.50 ± 1.88^{ab}	6.55 ± 1.47^{ab}
	14	6.05 ± 1.67^{b}	6.50 ± 1.64^{ab}	5.80 ± 2.28^{b}	6.35 ± 1.73^{b}
	21	5.75 ± 1.16^{c}	5.85 ± 1.95^{c}	5.40 ± 1.98^{d}	6.00 ± 1.52^{bc}
	28	5.10 ± 1.74^{d}	5.30 ± 2.00^{d}	5.00 ± 1.34^{d}	5.55 ± 1.96^{cd}

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

With regards to the overall acceptability, a decreasing trend was recorded in pit and *chorkor* smoked *M. caschive* and *O. niloticus* over storage time (Table 8). Low average acceptability scores were recorded on the last day than on the first day of the sensory evaluation. Concerning technologies, *chorkor* smoked fish were highly accepted than pit smoked fish. The overall acceptability means scores in pit and *chorkor* smoked *M. caschive* and *O. niloticus* were above 5, the critical limit for products under quality assessment.

Generally, evaluation of sensory attributes in pit and *chorkor* smoked *M. caschive* and *O. niloticus* during 28th days of storage depicted that smoked fish samples from *chorkor* technology were of better quality. Besides, *chorkor* smoked fish have higher mean scores in all the attributes than pit smoked fish products.

Relationship between water activity and microbial growth with the sensory attributes of smoked fish stored at ambient temperature

Pearson correlation results showed an inverse relationship between water activity and sensory characteristics with a range of -0.66 to -0.77 for the attributes. The lowest relationship was between water

Table 7. Trend in textural mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at ambient temperature.

Parameter	Storage time (Days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Texture	0	7.00 ± 1.56[a]	7.35 ± 1.87[a]	6.55 ± 2.01[ab]	7.00 ± 1.45[a]
	7	6.60 ± 1.19[a]	6.65 ± 1.42[a]	6.30 ± 1.56[b]	6.70 ± 1.78[a]
	14	5.60 ± 1.85[bc]	6.20 ± 1.58[b]	6.10 ± 1.77[b]	6.55 ± 2.26[ab]
	21	5.45 ± 1.54[c]	6.05 ± 1.28[b]	5.55 ± 2.04[bc]	6.35 ± 1.63[b]
	28	5.05 ± 1.88[c]	5.30 ± 1.72[c]	5.45 ± 1.99[c]	5.50 ± 1.47[bc]

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

Table 8. Trend in acceptability mean scores recorded in Pit and *Chorkor* smoked fish products stored for 28 days at ambient temperature.

Parameter	Storage time (days)	*M. caschive*		*O. niloticus*	
		Pit	*Chorkor*	Pit	*Chorkor*
Acceptability	0	6.80 ± 1.64[a]	7.15 ± 1.46[a]	7.05 ± 1.36a	7.25 ± 0.97[a]
	7	6.40 ± 1.54[b]	7.00 ± 1.75[a]	6.80 ± 1.54[a]	6.95 ± 1.50[a]
	14	6.20 ± 1.36[b]	6.85 ± 1.31[a]	6.65 ± 1.76[ab]	6.70 ± 1.45[a]
	21	5.35 ± 1.04[c]	6.50 ± 1.54[ab]	6.45 ± 1.54[b]	6.60 ± 1.19[ab]
	28	5.15 ± 1.69[c]	5.75 ± 1.80[b]	6.25 ± 1.02[b]	6.30 ± 1.42[b]

Mean with different superscript in the same column for each parameter are significantly different (P<0.05). SD=Standard deviation, Means ±SD are values of duplicate evaluation.

activity and appearance (r = -0.68), and highest between water activity and taste (r = -0.77). As water activity increases, the sensory quality of smoked fish reduces. Similarly, an indirect relationship between microbial load and sensory characteristics with a range of -0.70 to -0.81 was established for the attributes. The lowest relationship was between microbial growth and aroma (r = -0.70), and highest between microbial growth and taste (r = -81). As microbial load increases the sensory characteristics of smoked fish reduces, a reason for reduced acceptability of smoked fish during storage. Conversely, a direct and positive relationship between water activity and microbial load was established. As water activity increases, microbial load in smoked fish increases during storage with correlation value of 0.95. Equally, a positive and direct relationship between sensory characteristics of smoked fish stored at room temperature was established with a correlation range of 0.68 to 0.94 for the sensory attributes. The lowest relationship was between appearance and aroma (r = 0.68), and highest between appearance and taste (r = 0.94), and aroma and acceptability (r = 0.94).

DISCUSSION

Water activity in the muscles of smoked *M. caschive* and *O. niloticus*

Results of this study revealed that, the initial water activity in pit smoked fish was higher than recorded in *chorkor* smoked products. This is attributed to inefficient smoking that could not effectively remove water from fish tissues due to difficulties in controlling smoke heat and oxygen during the smoking process, given the openness of pit kiln. During storage, water activity increased with increase in storage time due to exposure of smoked fish to atmospheric air in the storage room. The rate of increase was higher in pit than in *chorkor* smoked fish samples due to higher initial water activity recorded in pit smoked fish (Ayeloja et al., 2018). Higher water activity allows microorganisms to infest the cracks, surfaces and the cavities or openings that subsequently digest smoked fish tissues.

Microbial load in the muscles of smoked *M. caschive* and *O. niloticus*

Consistent with Udochukwu et al. (2016) and Yusuf et al. (2017), the study recorded higher microbial count in fresh fish than in smoked fish samples. This could be attributed to unhygienic handling of fish at the landing site and during transportation (Amegovu et al., 2017; Baniga et al., 2017; Muhame et al., 2020). It could also be due to the fact that, the ice used for preserving the fresh fish melted during transportation owing to temperature differences. Indeed, microorganisms increased exponentially with increase in ambient temperature

(Moneim et al., 2012; Pal et al., 2016). Concerning smoking technologies, *chorkor* smoked fish recorded lower microbial count than pit smoked fish. This agrees with earlier studies (Nunoo and Kombat, 2013; Agu et al., 2013; Adeyeye et al., 2015). Low microbial load could be attributed to proper handling of fresh fish for smoking and sanitary *chorkor* facility (Osvaldo, 2016), effect of smoke chemicals and heat in preventing or reducing the proliferation capacity of microorganisms (Adeyeye et al., 2015). In pit kiln, fish smoking is done in an open and unhygienic environment, hence the chances for microbial contamination (Adelaja et al., 2013; Adeyemi et al., 2013; Omodara et al., 2016; Amegovu et al., 2017).

In relation to storage, results revealed an increasing microbial load in smoked fish with storage time. This observation is in line with the findings of Kumolu-Johnson et al. (2010) who reported an increase in microbial load in smoked *Clarias gariepinus* during storage and attributed it to storage temperature and duration. Ayinsa and Maalekuu (2013), Ayuba et al. (2013) and Olatunde et al. (2013) reported that the quality of smoked fish deteriorates with time during storage at ambient temperature. The decline was attributed to increase in microbial load associated with increase in moisture content absorbed from the atmosphere (Daniel et al., 2013; Udochukwu et al., 2016). Relative increase in microbial load of smoked fish stored at room temperature is an important indicator of quality, and influences the time in which smoked fish can be stored (Kumolu-Johnson et al., 2010; Adeyemi et al., 2013). In the present study, increase in water activity due to moisture reabsorption during storage corresponded with increase in microbial load. This supports the observation that microbial load correlates well with water activity and storage time (Ayuba et al., 2013). Indeed, dehydrated fish easily absorbs water from the surrounding air into their tissues triggering microbial growth with subsequent increase in the load (Daniel et al., 2013). Despite the increase of microbes in smoked fish, the loads were below the maximum permissible limits recommended by the International Standards (1.0×10^6 and 1.0×10^4 cfu/g) for bacteria and fungi, respectively.

Similarly, the study recorded higher mould count in fish smoked using pit than *chorkor*, and the load increased during storage at ambient temperature. This could be due to poor processing techniques employed in traditional pit kiln (Sajib et al., 2015) and exposure of the products during storage (Udochukwu et al., 2016). In addition, the findings showed moulds are capable of growing at water activity of 0.50. This is in line with Daniel et al. (2013) who revealed that moulds are the major microorganisms thriving in smoked and dried fish products with low water activity (0.60). Concerning microbial load and storage, it is imperative to say that pit smoked fish has low shelf life than *chorkor* smoked fish and consumption of such fish after 35th day of storage may result to potential health effects (Udochukwu et al., 2016).

Sensory evaluation of organoleptic attributes of smoked fish products

The overall results for sensory evaluation (Figure 5a and b) showed higher scores in *chorkor* smoked *M. caschive* and *O. niloticus* than pit smoked fish. With regards to the technologies, *chorkor* products had higher colour mean scores than pit smoked samples. Unlike the black and unattractive colour observed from pit smoked fish, *chorkor* products had attractive dark-brown colour. This could be due to the difference in proximity of fish to heat source, deposition and adsorption rate of smoke chemicals by fish surface during smoking. The colour difference could also be due to variation in smoking parameters that is, time and temperature as fish samples from pit were directly exposed to intense smoke heat for a longer period (18-24 h) than products from *chorkor* kiln (6-12 h). Besides, the difference could be due to the effect of oxidation or rancidity that might have occurred during smoking and storage. Studies observed that large amount of polyunsaturated fatty acids found in fish lipids make smoked fish liable to oxidation and autocatalytic reactions which may lead to production of higher peroxides during storage (Nguvava, 2013; Olukayode and Paulina, 2017).

In relation to taste, *chorkor* smoked *M. caschive* and *O. niloticus* have better taste scores than pit smoked fish. This could be due to salting effect and regulated smoking events which might have prevented fish products from contamination with microorganisms, external materials and smoke chemicals (Likongwe et al., 2018). Generally, the taste of smoked fish deteriorated during storage at ambient temperature with higher rate of taste decline recorded in pit smoked fish than in *chorkor* smoked products. This could be attributed to variation in smoke concentration which could have resulted to fish contamination by organic pollutants (Dutta et al., 2018), and difference in salt absorption from the surface into fish muscles. Besides, increase in water activity increases microbial load, and this reduces the taste of smoked fish during storage (Adeyeye et al., 2015). The rate of taste reduction differed between pit and *chorkor* smoked fish which is depending on the amount of water activity that directly influences microbial growth (Pal et al., 2016).

With regards to aroma, *chorkor* smoked fish had higher aroma mean scores than pit smoked products but a general decline in the pleasant aroma of fish was noted during storage. This difference could probably be due to variation in the rate of fat and protein decomposition due to heat treatment and subsequently, oxidation of poly-unsaturated fatty acids (PUFA) contained in fish muscles to products such as peroxides, aldehydes, ketones and free fatty acids during storage at ambient temperature (Olatunde et al., 2013; Stratev et al., 2015; Abdul-Baten et al., 2020). The difference in aroma could also be attributed to decomposition of nutrients by microorganisms during storage (Adeyemi et al., 2013;

Famurewa et al., 2017). This is evidenced by the inverse relationship between water activity and microbial growth with aroma. As water activity increases, microbial load also increases that reduced the sensory characteristics of smoked fish particularly aroma.

For flavour, *chorkor* smoked fish had better mean scores than pit smoked fish but a general decline in score was recorded in all smoked fish products during storage. Pre-treatment of fish with salt prior to smoking could have caused the difference as salt contributes to development of aroma and flavour (Odoli, 2015). Reduction in flavour during storage could probably be due to absorption of water owing to difference in osmotic pressure which existed between salted smoked fish and humidity. Moisture adsorbed increases water activity which directly influenced microbial growth (Pal et al., 2016). Due to increase in water activity, microorganisms are enabled to decompose proteins and fats to products such as amines, ammonia, peroxides and free fatty acids that may result to off flavour and rancid taste. This supports Famurewa et al. (2017) observation that fish nutrients are metabolized and reduced as spoilage parameters increases during storage.

In terms of texture, fish products smoked in *chorkor* were highly preferred by the panelists to pit smoked samples. This could be due to low water activity, and application of salt that retarded the activities of spoiling microorganisms and other chemical reactions (Reza et al., 2015), which would have contributed to undesirable textural changes. In line with other studies (Saritha et al., 2012; Adeyeye et al., 2015; Jakhar et al., 2015), variation in the overall acceptability of smoked fish could be attributed to efficacy of the technology in removing water that has an impact on visual and nutritive quality of smoked fish (Abdul-Baten et al., 2020).

The overall acceptability was higher for fish smoked using *chorkor* than pit. This is because the temperature used for smoking fish was not excessively high (60-80°C) as it was reasonably controlled. This has made most of the physical and nutritive qualities to be retained at the end of the smoking process, giving rise to products which were very pleasant, delicious and attractive as judged by the panelists. During storage, an inverse relationship between water activity and microbial growth with acceptability of smoked fish was established. As water activity increases so is microbial load that reduced the acceptability of smoked fish (Dutta et al., 2018).

On average, all fish smoked using pit and *chorkor* technologies were acceptable 28th days after smoking. This could be due to curing of fresh fish with salt prior to smoking that improves the taste, palatability and flavour (Magawata and Musa, 2015; Odoli, 2015; Ginigaddarage et al., 2018). Most importantly, salting of fish before smoking reduces and controls the activities of microorganisms (Chakraborty and Chakraborty, 2017) which may otherwise metabolize proteins, fats and poly-unsaturated fatty acids (PUFA) in fish muscle to oxidation products such as amines, ammonia, peroxides, aldehydes, ketones and free fatty acids during storage at ambient temperature (Ayeloja et al., 2018) which may cause changes in colour, aroma, flavour and texture of fish (Olatunde et al., 2013).

Most important to note is that, in all the sensory attributes examined, *chorkor* smoked fish scored higher than pit. However, all smoked fish scored above average, which indicates that pit and *chorkor* smoked fish might still be appealing and acceptable beyond a period of one month after smoking. However, the microbial load needs to be taken into account. In agreement with earlier studies (Daniel et al., 2013; Odoli, 2015; Ginigaddarage et al., 2018), fish smoked using *chorkor* might have longer shelf life than fish smoked using pit due to low water activity, low microbial load, and good sensory characteristics as judged by the panelists.

Conclusion

The study revealed that *chorkor* removes water in fish more significantly than pit. It is also associated with low microbial load and good quality sensory parameters. Regardless of smoking technology, fish tissues continuously absorb atmospheric moisture during ambient storage resulting in increased water activity that led to increase in microbial loads. Despite rise in microbial load, fish were safe for human consumption as the loads were below the maximum permissible limits specified by the International Standards. The study recommends adoption of improved *chorkor* characterized with easy control of smoking parameters and maintenance of smoked fish quality. Uptake of improved *chorkor* will consolidate smoked fish value chain by maintaining the quality and quantity of smoked fish products that will enhance the role of fish in providing food and nutrition security in South Sudan, Africa.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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