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# Production and Physicochemical Evaluation of Lemon Grass and Bay Leaf Extract as Natural Preservatives in Smoked African Catfish

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## Abstract

This study evaluated the effect of lemongrass (Cymbopogon citratus) and bay leaf (Laurus nobilis) extracts, individually and in combination, on the shelf stability of smoked African catfish (Clarias gariepinus) during 28 days of storage. The treatments were assessed for changes in water activity, thiobarbituric acid (TBA), peroxide value (PV), free fatty acids (FFA), total viable counts (TVC), and mould growth. Results showed that water activity, TBA, PV, and FFA increased progressively across all samples during storage. However, fish treated with the combination of lemongrass and bay leaf consistently exhibited significantly lower values ($p < 0.05$) compared to the untreated control. Similarly, microbial and mould growth were markedly reduced in the extract-treated groups, with the combined treatment being most effective. The findings demonstrate that lemongrass and bay leaf extracts, especially when applied together, can effectively retard lipid oxidation and microbial proliferation, thereby improving the storage quality and extending the shelf life of smoked catfish.

**Keywords:** African catfish, lemongrass, bay leaf, preservatives, shelf life, microbial quality

## Introduction

Fish and fishery products represent an important source of high-quality protein, essential fatty acids, vitamins, and minerals for millions of people worldwide (FAO, 2022). In many developing countries, including Nigeria, fish is a staple in household diets and contributes significantly to food and nutritional security (Obiero *et al*., 2019). However, fresh fish is highly perishable due to its high moisture content, enzymatic activity, and susceptibility to microbial spoilage, which often lead to deterioration in sensory and nutritional quality within a short period after harvest (Shahidi and Zhong, 2015; Fasuan *et al*., 2022). Most consumers of fish in Nigeria benefit from fish smoking for its taste and aroma with longer shelf life as a result of the combined effects of dehydration, antimicrobial and antioxidant activities of several smoke constituents such as formaldehyde, makes it an important fish processing technique in the Nigerian traditional diet (Famurewa *et al.,* 2017).

To address this, fish is often subjected to traditional preservation techniques such as smoking, salting, and drying. Smoking remains one of the most common preservation methods in sub-Saharan Africa because it not only improves flavor but also reduces moisture content, thereby extending shelf life (Chukwu *et al*., 2023). Despite this advantage, smoked fish is still vulnerable to lipid oxidation, microbial contamination, and mould growth during storage, particularly under tropical conditions where temperature and humidity accelerate spoilage processes (Ndife *et al*., 2022).

Microbial proliferation is another major concern for smoked fish products. While smoking reduces microbial load, spores of bacteria and fungi often survive and grow under favorable storage conditions. Common contaminants include *Bacillus*, *Pseudomonas*, and mould species such as *Aspergillus* and *Penicillium* (Obiero *et al*., 2019). High microbial loads compromise food safety and reduce consumer acceptability. Codex Alimentarius (2020) recommends that smoked fish products maintain microbial loads below $10^5$ cfu/g during storage to ensure safety. Lipid oxidation in fish products produces undesirable compounds such as aldehydes and ketones that cause rancid off-flavors, while microbial growth contributes to reduced safety and marketability (Eymard *et al*., 2019). The use of synthetic antioxidants and preservatives has been practiced in the food industry; however, consumer demand is shifting toward natural, plant-based alternatives due to concerns over the safety of chemical additives (Shahidi and Ambigaipalan, 2015).

Starting from the food preparation, plant-derived natural compounds can affect both food spoilage microorganisms (food preservation) and human pathogens (food safety) due to the antimicrobial and antifungal activity of their natural constituents (Anamaria *et al.,* 2019). Due to increasing consumer demand for natural food systems, plant-based extracts and essential oils are being explored as alternatives to synthetic preservatives (Shahidi and Ambigaipalan, 2015). Lemongrass (*Cymbopogon citratus*) is rich in citral, geraniol, and flavonoids, compounds known for their antimicrobial and antioxidant activities (Omoruyi *et al*., 2019). Bay leaf (*Laurus nobilis*) contains eugenol, linalool, and phenolic acids, which exhibit strong radical scavenging and microbial inhibitory effects (Akinmoladun *et al*., 2021). It is a common household spice used in flavoring a wide range of foods and possesses several medicinal properties. (Raman *et al.*, 2017). Previous studies have shown that these plant extracts can retard lipid oxidation and inhibit microbial growth in various food systems (Obiero *et al*., 2019). Natural preservatives are available from a variety of sources including plants, animals, bacteria, algae, and fungi. (Busra and Gulsun, 2021). However, their application in smoked fish, particularly African catfish (*Clarias gariepinus*), has not been extensively studied.

Therefore, this study investigated the effects of lemongrass and bay leaf extracts, individually and in combination, on the chemical and microbiological stability of smoked catfish during storage. By evaluating parameters such as water activity, lipid oxidation indices (TBA, PV, FFA), microbial load, and mould counts**,** this research aims to establish the potential of natural extracts as sustainable preservatives in smoked fish processing.

## Study Materials

The raw materials were African Catfish, a total of 24 fishes with a total weight of 36 kg, Lemon grass (30g) and Bay leaves (30g) used for this project were purchased from Ogige market in Nsukka town. Each sample consisting of two African catfish weighing

Catfish (2 per sample) weighing 1.5kg each were used for both the control and treated samples. The Catfish samples were stunned, gutted and washed. After which they were brined by dipping into 75% saturated brine solution made by dissolving 25g of NaCl in 100ml of water for 3 minutes. The fishes were rinsed with cool clean water. Fresh lemon grass was selected, washed thoroughly to remove impurities and cut into thin small slices. Then 30g of sliced lemon grass was put in a blender and 100ml of water was added to the blender to obtain the marinade.

The Bay leaves were sorted and 30g of bay leaves was weighed using an electric weighing balance, then washed thoroughly, after which the bay leaves were blended with 100ml of water using a blender with high speed to obtain the bay leaf marinade. The extract was used to marinate the fish samples for 20 minutes. The smoking kiln was heated and the fish smoked at a temperature range of 80°C - 100°C for 24 hours.

## Methodology

The Water activity (aW), thiobarbituric acid (TBA), peroxide value (PV) and free fatty acid value (FFA) of the fish samples were determined at 0, 14th and 28th days of storage using standard methods of AOAC (2010). The Total viable and mould count were determined by pour plate method described by (Prescott *et al*., 2005). Test samples were drawn at 0, 14th and 28th day of storage

## Results and Discussion

**Table 1:** Effect of Storage on the Water activities, Thiobarbituric acid (TBA), Peroxide value (PV) and Free fatty acids (FFA) of smoked catfish treated with lemongrass and bay leave.

| Samples | Storage days | Water activity (%) | TBA(mg MDA/kg) | PV(Meq$O_2$/kg) | FFA (%) |
|---|---|---|---|---|---|
| (CT) Control | | | | | |
| CT1 | Zero day | $0.73^{c}\pm0.01$ | $0.19^{d}\pm0.014$ | $5.35^{b}\pm0.14$ | $0.89^{b}\pm0.19$ |
| CT2 | 14th day | $0.78^{c}\pm0.01$ | $0.29^{d}\pm0.003$ | $10.33^{d}\pm0.22$ | $1.06^{cd}\pm0.07$ |
| CT3 | 28th day | $0.90^{d}\pm0.01$ | $0.35^{d}\pm0.006$ | $12.42^{e}\pm0.07$ | $1.39^{d}\pm0.02$ |
| (CL) Lemongrass | | | | | |
| CL1 | Zero day | $0.71^{b}\pm0.01$ | $0.16^{b}\pm0.003$ | $4.96^{b}\pm0.58$ | $0.84^{c}\pm0.08$ |
| CL2 | 14th day | $0.75^{b}\pm0.01$ | $0.25^{c}\pm0.001$ | $7.12^{c}\pm0.24$ | $0.98^{c}\pm0.05$ |
| CL3 | 28th day | $0.88^{c}\pm0.01$ | $0.33^{c}\pm0.004$ | $9.33^{d}\pm0.59$ | $1.36^{d}\pm0.03$ |
| (CB)Bay leave | | | | | |
| CB1 | Zero day | $0.71^{b}\pm0.01$ | $0.17^{c}\pm0.004$ | $4.22^{ab}\pm0.60$ | $0.61^{b}\pm0.07$ |
| CB2 | 14th day | $0.74^{b}\pm0.01$ | $0.21^{b}\pm0.002$ | $6.54^{b}\pm0.15$ | $1.20^{d}\pm0.07$ |
| CB3 | 28th day | $0.85^{b}\pm0.01$ | $0.28^{b}\pm0.001$ | $8.73^{c}\pm0.41$ | $1.77^{d}\pm0.05$ |
| (CBL)Bay leave+Lemongrass | | | | | |
| CBL1 | Zero day | $0.67^{a}\pm0.01$ | $0.10^{a}\pm0.002$ | $3.84^{a}\pm0.43$ | $0.36^{a}\pm0.05$ |
| CBL2 | 14th day | $0.70^{a}\pm0.01$ | $0.19^{a}\pm0.002$ | $5.21^{b}\pm0.19$ | $0.50^{a}\pm0.01$ |
| CBL3 | 28th day | $0.74^{a}\pm0.01$ | $0.20^{a}\pm0.002$ | $7.69^{c}\pm0.05$ | $0.77^{b}\pm0.04$ |

Values are means at ± standard deviation of duplicate determinations. Values bearing different superscripts within the same column are significantly ($p<0.05$) different

**Key:** Sample CBL (smoked fresh fish treated without lemongrass and bay leave CT1, CT2, CT3) = Control; Sample CL (treated with only lemongrass CL1, CL2, CL3); Sample CB (treated with only bay leave CB1, CB2, CB3); Sample CBL (treated with both lemongrass and bay leave CBL1, CBL2, CBL3)

The results of the aW, TBA, PV and FFA value of the fish samples treated with lemon grass and bay leave is shown in Table 1 During the storage period, a consistent increase in water activity was observed in all the samples. The water activity values increased from 0.67 to 0.90 with sample CT3 (Control sample at day 28 storage) having the highest value while sample CBL1 (Bayleaf and lemongrass treatment at day1) showed the lowest aw value. There was a significant difference in the aw levels of the fish samples ($p < 0.05$). Obiero et al., (2019) reported similar water activity values of smoked catfish samples of 0.70 and 0.79 for 30 days storage period. The increase in aW observed in the study could be due to exposure of the samples to atmospheric humidity during storage. Within the first 2 weeks of storage the water activity of all the samples was below the Codex Standard for smoked and oven-dried fish which is 0.75. This standard is necessary to control bacteria pathogens and spoilage fungi (Fasuan *et al.,* 2022). However, the water activity of the samples CT3, CL3, and CB3 at day 28 was above the Codex Standard for smoked fish. This points to the synergistic effect of bay leave and lemon grass treatment for sample CBL3. This can be as a result of antioxidant properties of lemongrass and bay leave which help to reduce water activity by binding to water molecules, thereby making it more difficult for the fish to absorb moisture from the surrounding as observed by Al-Hashimi and Mahmood (2016).

Thiobarbituric acid value (TBA) assesses how secondary lipid oxidation products, primarily malondialdehyde, are formed and may be responsible for the off-flavour of oxidized fat (Zhang *et al*., 2016). The TBA values for the smoked catfish treated with lemon grass and bay leave (CBL, CL and CB) were consistently lower than those of the control sample (CT1, CT2 and CT3) which was not treated with lemongrass and bay leave. The TBA values was highest in untreated sample (CT3) at the 28th day (0.35 mg MDA/kg) and lowest in the sample treated with both bay leave and lemon grass (CBL1) at the first day (0.10 mg MDA/kg). There was a significant difference in the TBA levels of the fish samples ($p < 0.05$). The low TBA values on the zero (0) day could probably be due to the destruction of pro-oxidants like lipase enzymes responsible for hydrolytic rancidity during smoking, drying and also due to anti-oxidative effect of phenol in lemongrass and bay leave (Nagawaka *et al*., 2007). All samples showed an increase in the TBA values during the storage period of 14$^{th}$ and 28$^{th}$ days. The TBA value increased with time. Generally, the untreated samples showed the higher TBA value than the samples treated with bay leave and lemon grass. The thiobarbituric acid levels found in this study were within the acceptable limit (3-4 mg MDA/kg) (Ndife *et al.,* 2022).

Peroxide value increased with storage period as shown in table 1. The PV ranged from 3.84 MeqO$_2$/kg to 12.42 MeqO$_2$/kg. It was highest in the untreated sample (CT3) and lowest in the sample treated with bay leave and lemon grass. There was a significant difference in the PV of the fish samples ($p < 0.05$). It was observed that the PV of the fish samples increased upon storage. The control sample (CT1) showed the highest peroxide value of 124.2 MeqO$_2$/kg while the sample treated with combination of bay leave and lemon grass showed the lowest peroxide value. Lower peroxide values were reported by Olatunde *et al*. (2022) (0.21 to 2.85 mEqO$_2$/kg) for smoked catfish treated with a mixture of garlic, ginger and turmeric during a 20 days storage period. These findings about the impact of antioxidants from bay leave and lemon grass on fish samples are consistent with the trend that Ihuahi *et al*. (2007) reported for stored smoked catfish in which 5% garlic and black pepper paste gave the lowest peroxide value for a period of 21days storage. The peroxide values of the treated samples recorded in this finding were within the permissible limit (below 10 mE qO$_2$ /kg) indicating that the products will keep well during storage (Ndife *et al.,* 2022), while the untreated samples exceeded the acceptable limit.

Changes in free fatty acid of smoked catfish treated with lemon grass and bay leave during storage are presented in table 1. The result revealed that the FFA values of the smoked fish increased upon storage. Control sample (CT3) recorded the highest FFA value of 1.39% while the sample treated with lemongrass and bay leave (CBL) had the lowest value of 0.36% at zero day. This suggests that FFA might have originated from partial hydrolysis of phospholipids during drying. Free fatty acids arise from hydrolysis of triacyl-glycerides in the presence of heat and water (Nakagawa *et al*., 2007). The FFA value of the sample control (CT1, CT2 and CT3) increased from 0.89 to 1.39%, samples treated with lemongrass (CL1, CL2 and CL3) increased from 0.84 to 1.36%, samples treated with bay leave (CB1, CB2 and CB3) 0.61 to 1.77% and samples treated with lemongrass and bay leave (CBL1, CBL2 and CBL3) 0.36 to 0.77%. This is in agreement with (Eyamard *et al*., 2019) who reported that storage increased the free fatty acid of horse mackerel minced fish.

**Table 2:** Total viable count (cfu/g) and Mould count (cfu/g) of smoked catfish treated with lemongrass and bay leave during storage.

| Samples | Storage days | TVC (cfu/g) | Mould count (cfu/g) |
|---|---|---|---|
| (CT) Control | | | |
| CT1 | Zero day | $3.5\times10^{1}$ | ND |
| CT2 | $14^{th}$ day | $7.0\times10^{4}$ | $6.0\times10^{3}$ |
| CT3 | $28^{th}$ day | $9.5\times10^{5}$ | $8.8\times10^{3}$ |
| (CL) Lemongrass | | | |
| CL1 | Zero day | ND | ND |
| CL2 | $14^{th}$ day | $1.7\times10^{4}$ | $4.5\times10^{3}$ |
| CL3 | $28^{th}$ day | $3.3\times10^{5}$ | $6.6\times10^{3}$ |
| (CB) Bay leave | | | |
| CB1 | Zero day | ND | ND |
| CB2 | $14^{th}$ day | $1.3\times10^{4}$ | $3.1\times10^{3}$ |
| CB3 | $28^{th}$ day | $2.9\times10^{5}$ | $4.7\times10^{3}$ |
| (CBL) Bayleave+ Lemongrass | | | |
| CBL1 | Zero day | ND | ND |
| CBL2 | $14^{th}$ day | $1.0 \times10^{4}$ | $2.5\times10^{3}$ |
| CBL3 | $28^{th}$ day | $2.3\times10^{5}$ | $3.3\times10^{3}$ |

**Key:** ND (Non detected); Sample CT (smoked fresh fish without lemongrass and bay leave CT1, CT2, CT3) = Control; Sample CL (treated with only lemongrass CL1, CL2, CL3); Sample CB (treated with only bay leave CB1, CB2, CB3); Sample CBL (treated with both lemongrass and bay leave CBL1, CBL2, CBL3)

The mould and total viable count of the smoked African Catfish treated with Lemon grass and Bay leaf extract. The total viable and mould count of the smoked catfish treated with lemon grass and bay leaf extract was evaluated. It was observed that during storage, there was more rapid proliferation of microorganisms in the control sample CT (without lemongrass and bay leave) as shown in table 1. The samples treated with the plant material extract showed lower microbial growth. As storage period advanced, the total microbial counts were similar for the samples treated with lemongrass and the samples treated with bay leave, with the samples (CL1, CB1 and CBL1) maintaining lower counts. TVC was not detected in the treated samples at day one but increased upon storage. The result shows that microbial growth was highest in the control sample CT2 and CT3 ($7.0x10^{4}$ cfu/g and $9.5x10^{5}$ cfu/g) on the $14^{th}$ day and $28^{th}$ days. Lower growth was recorded in the samples (CL2, CB2 and CBL2) $1.7x10^{4}$ cfu/g, $1.3x10^{4}$ cfu/g and $1.0x10^{4}$ cfu/g respectively on the $14^{th}$ day. Synergistic antimicrobial and antioxidant effect of lemon grass and bay leave was observed to lower growth rate of microorganisms in this study. Storage significantly increased the total viable count, in line with the observation made by (Omoruyi *et al*., 2019), who reported that microbial load increases with duration of storage.

From table 2, it was noted that mould count on the various smoked fish samples increased with the storage period. No mould count was observed at zero day. The mould count was highest in the untreated samples at 14 and 28 days (CT2 and CT3) and also in the samples treated with bay leaf and lemon grass at the 28 day (CB3 and CL3). This may be due to the presence of phenols and other bioactive compounds in treated sample (CB, CL and CBL) with the samples treated with both bay leave and lemon grass having the least mould count. Similar observation has been reported by (Egbal *et al*., 2010) in salted kass fish stored at room temperature (37±1°C). This delayed proliferation of the microorganisms may also be linked to decreased moisture content, as fungi are saprophytic organisms that thrive in moist environments, thus reduced water activity limits their proliferation (Omoruyi *et al*., 2019; Chukwu *et al*., 2023). Recent studies confirm that plant-derived preservatives such as extracts of Moringa (*Moringa oleifera*), lemongrass (*Cymbopogon citratus*), and bay leaf (*Laurus nobilis*) exhibit significant antimicrobial activity against common foodborne spoilage microorganisms (Abdallah *et al*., 2023).

These activities have been demonstrated for both chloroform and ethanol extracts, highlighting their potential as natural sanitizers and shelf-life extenders in food preservation (Akintelu *et al*., 2021).

## Conclusion

The findings of this study demonstrated that the application of lemongrass (*Cymbopogon citratus*) and bay leaf (*Laurus nobilis*) extracts, individually and in combination, effectively improved the storage stability and quality of smoked African catfish (*Clarias gariepinus*). Samples treated with both plant extracts (CB, CL and CBL) exhibited significantly lower water activity, thiobarbituric acid values, peroxide values, free fatty acids, total viable counts, and mould counts compared to the control, indicating reduced lipid oxidation and microbial proliferation during the 28-day storage period. The synergistic effect of lemongrass and bay leaf was particularly evident in maintaining superior microbiological quality and delaying oxidative spoilage, thereby extending shelf life under ambient conditions. These results underscore the potential of natural plant extracts as safe, affordable, and consumer-acceptable bio-preservatives in smoked fish processing. Adopting such natural preservation strategies can reduce post-harvest losses, enhance product safety, and improve the economic returns for fish processors, while meeting the growing consumer demand for minimally processed foods free from synthetic additives.

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