International Journal of Science and Technology (IJST)
Vol. 9:000003 (2025) 2507-9638 https://ijst.clsu.edu.ph

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Effects of Different Smoking Materials on the Microbiological, Polycyclic Aromatic Hydrocarbons, and Sensory Characteristics of Milkfish (*Chanos chanos*)

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Keywords: *Coconut shells, Mango wood shavings, Microbial Count, Milkfish processing techniques, Polycyclic aromatic hydrocarbon, Santol wood shavings*

Type: Research Article

Submitted: March 14, 2025
Accepted: December 15, 2025
Published: December 31, 2025

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Citation
Ogdamin, D., Villota, E., Mateo, W., Sayco, T., & Badua, S. (2025). Effects of Different Smoking Materials on the Microbiological, Polycyclic Aromatic Hydrocarbons, and Sensory Characteristics of Milkfish (*Chanos chanos*). *CLSU International Journal of Science and Technology, 9*, 000003. https://doi.org/10.22137/IJST.2025.000003

Abstract

Smoked fish is a staple in many traditional dishes, yet awareness of its potential health risks, particularly concerning polycyclic aromatic hydrocarbons (PAHs) and microbial contamination, remains limited, particularly in rural communities of Capiz, Philippines. To date, there is insufficient information on polycyclic aromatic hydrocarbon (PAH) levels associated with different types of wood used in smoking. This study aimed to evaluate the effects of three smoking materials—coconut (*Cocos nucifera)* shell (CCS), mango (*Mangifera indica)* wood shavings (MWS), and santol *(Sandoricum koetjape*) wood shavings (SWS)—on the sensory attributes, PAH levels, and microbial load of smoked milkfish (*Chanos chanos*). Eighteen fish samples were smoked per treatment, with nine randomly selected for PAH and microbial analyses. Results showed that PAH levels in all samples were below the detection limit (<10 µg/kg), confirming compliance with food safety standards. Microbial analysis revealed significantly lower counts in CCS-smoked fish (370 CFU/g) compared to MWS and SWS smoked samples, which exceeded 6,500 CFU/g but remained well below the Food and Drug Administration (FDA) maximum allowable limit of 5×10^5 CFU/g. Sensory evaluation indicated a strong preference for MWS-smoked fish due to its aroma, taste, and mouthfeel. These findings highlight the potential of CCS for reducing microbial contamination, while MWS enhances sensory attributes, making it suitable for premium smoked fish products. This study provides insights into optimizing smoking processes to ensure food safety and improve consumer acceptability of smoked milkfish.

Introduction

The milkfish (*Chanos chanos*) industry is a cornerstone of the Philippine aquaculture sector, significantly contributing to the nation's economy. In Capiz, a province renowned for its aquaculture activities, milkfish farming plays a vital role in sustaining local livelihoods and ensuring food security. As of the second quarter of 2025, milkfish is among the top aquaculture products in the Philippines, with a production volume of 77.23 thousand metric tons, accounting for 7.8 percent of the country's total fisheries production (Philippine Statistics Authority [PSA, 2025]). Given the increased production of milkfish, it is critical to implement safe processing methods to ensure its preservation, maintain quality, and extend shelf life while preventing post-harvest losses.

Smoking is a widely used preservation method in the Philippines, valued for imparting desirable sensory attributes and extending shelf life. However, the process, particularly the type of smoking material and conditions used, can introduce harmful compounds such as polycyclic aromatic hydrocarbons, which are potentially carcinogenic and mutagenic (Hamidi *et al.*, 2016; Mičulis *et al.*, 2011; Çiftçi & Ayas, 2021). Several studies have reported the presence of these compounds in smoked fish (Joseph *et al.*, 2021; Sampaio *et al.*, 2021; Kafeelah *et al.*, 2015), as PAHs are naturally formed during the incomplete combustion and pyrolysis of organic materials, including the wood used in smoking (Joseph *et al.*, 2021). The formation mechanism involves the thermal degradation of lignocellulosic biomass, composed primarily of cellulose, hemicellulose, and lignin (Chen *et al.*, 2022). During pyrolysis, these components decompose, with different constituents yielding various volatile species. For instance, cellulose and hemicellulose contribute carbohydrates and aldehydes, while lignin produces phenolic compounds. These pyrolysis products can serve as crucial precursors for PAH and soot formation, particularly at higher temperatures and under conditions of incomplete combustion (Chen *et al.*, 2022; Matamba *et al.*, 2021). The specific composition of the smoking material and the control over the combustion process are thus critical in mitigating the formation of these contaminants (Matamba *et al.*, 2021; Savin *et al.*, 2024).

Concerns over food safety, especially regarding such carcinogenic compounds, have led to the establishment of stringent international regulations. The European Union has set comprehensive limits for PAHs in food products. Specifically, EC Regulation No. 835/2011 (amending Regulation No 1881/2006) mandates maximum levels for key PAHs in various foodstuffs, including smoked fish (Catena *et al.*, 2020; Racoviţă *et al.*, 2021). This regulation stipulates a maximum level of 5 μg kg^{-1} for Benzo[a]pyrene and a combined limit of 30 μg kg^{-1} for the sum of four specific PAHs (PAH4: Benzo[a]pyrene, Benzo[a]anthracene, Chrysene, and Benzo[b]fluoranthene) in smoked meat and fish products (Catena *et al.*, 2020; Racoviţă *et al.*, 2021). These international standards serve as vital benchmarks for assessing the safety of smoked products and underscore the global imperative to adopt processing methods that minimize PAH contamination.

The challenge of PAH contamination in smoked fish is a widespread concern, particularly prevalent in tropical countries where traditional smoking methods are essential for food preservation and economic activity. Studies from various regions, including Benin, Ghana, and Togo, consistently report high levels of PAHs in traditionally smoked fish, often exceeding international safety limits and posing significant health risks to consumers (Assogba *et al.*, 2024; Hasselberg *et al.*, 2020; Joseph *et al.*, 2021). These studies frequently highlight that traditional processing techniques, characterized by direct product exposure to smoke, high combustion chamber temperatures, and specific fuel types, contribute to substantial PAH contamination (Asamoah *et al.*, 2021; Assogba *et al.*, 2024; Assogba *et al.*, 2022). The commonality of these issues across diverse tropical settings underscores the global relevance of research into optimizing smoking practices to ensure both product quality and consumer safety, including within the Philippine context.

Beyond PAH formation, smoke also plays a crucial role in the microbial safety of food. Wood smoke is a complex mixture containing various chemical compounds, including phenols, organic acids, and carbonyls, which are generated from the thermal breakdown (pyrolysis) of wood components (Savin *et al.*, 2024; Ekonomou *et al.*, 2020). These compounds exert antimicrobial effects by damaging bacterial membranes and inactivating essential enzymes, thereby inhibiting the growth of spoilage and pathogenic microorganisms and contributing to the preservation of smoked products (Chen *et al.*, 2022; Ekonomou *et al.*, 2020) Furthermore, the specific types and concentrations of volatile phenolics released during smoking are dependent on the type of wood used, highlighting the critical influence of smoking material composition on the antimicrobial efficacy of the smoke. (Albishi *et al.*, 2019).

The quality and safety of smoked fish are influenced by various factors, including the type of wood used, salting or brining methods, processing conditions, and storage practices (Speranza *et al.*, 2021; Ali *et al.*, 2022; Abolagba & Nuntah, 2011). In Capiz, traditional smoking materials such as guava, jackfruit, and mahogany have become scarce due to environmental changes, prompting fish processors to explore locally available alternatives like santol (*Sandoricum koetjape)* wood shavings, mango (*Mangifera indica)* wood shavings, and coconut (*Cocos nucifera)* shells. Despite their local availability and potential use, there is a notable lack of scientific data on their specific effects on smoked milkfish quality, PAH formation, and microbial safety. This gap limits informed decision-making regarding safe and sustainable smoking practices in the region. Therefore, this study aimed to establish safe processing methods for smoked milkfish using these locally available smoking materials. Specifically, this study aimed to: Determine the Polycyclic Aromatic Hydrocarbon levels in smoked milkfish associated with each smoking material using Gas Chromatography Mass Spectrometry. Determine the microbial load of smoked milkfish processed with each smoking material using the Standard Plate Count method and evaluate the effect of santol wood shavings, mango wood shavings, and coconut shells on the sensory characteristics (aroma, color, taste, and overall acceptability) of smoked milkfish using 85 consumer panelists and a 9-point hedonic scale.

Materials and Methods

Ethical Approval

Ethical approval for the use of milkfish (*Chanos chanos*) in this research was obtained and approved by the Chair of the ethics review committee under approval number 2023-631 at Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines. For the human sensory evaluation component, all participants provided informed written consent before their involvement in the study, adhering to ethical research practices.

Pre-smoking Preparation

The transportation, housing, and overall welfare of the laboratory fish adhered strictly to the standard operating procedures outlined in the university's guidelines and regulations. Careful handling was maintained throughout the study in compliance with the Animal Research: Reporting of In Vivo Experiments guidelines to ensure ethical research practices.

The smoking process was conducted at the Technology Business Incubator, Central Luzon State University, Science City of Muñoz, Nueva Ecija. A total of 20.55 kg of fresh milkfish was used, along with 6.5 kg each of coconut (*Cocos nucifera*) shells, mango (*Mangifera indica)* wood shavings, and santol (*Sandoricum koetjape)* wood shavings. The wood shavings were obtained from branches cut five days before use. The bark was removed from the wood before smoking to prevent the introduction of potentially undesirable compounds that could affect the flavor and safety of the smoked fish. All smoking materials were sourced from local markets.

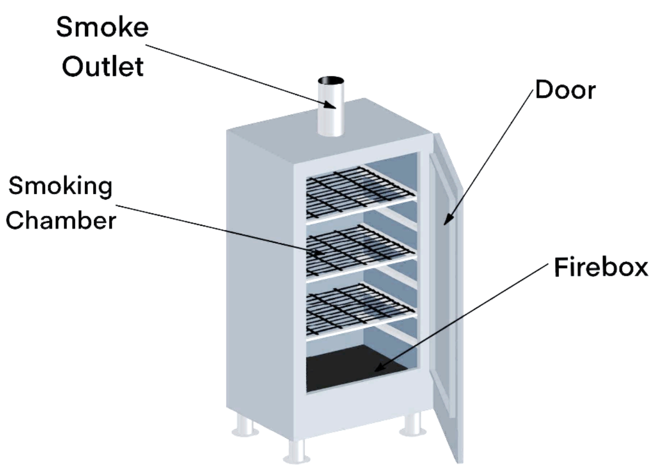

Figure 1. Traditional Smokehouse used in the study

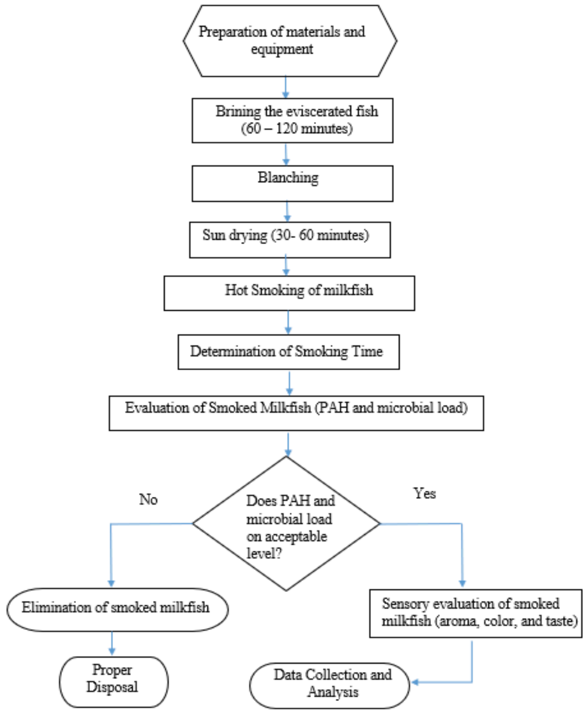

Figure 2. A schematic diagram of the experimental workflow.

The fish samples were smoked using a traditional smokehouse manufactured by Meat Packers and Butchers Supply Corporation, Los Angeles, California. This closed cubic metal smokehouse, with a 30 kg fish capacity, consists of three functional sections (Figure 1). The study workflow outlining the preparation, smoking procedures, and evaluation of the smoked milkfish samples is shown in Figure 2.

Preparation of Milkfish Samples

Live milkfish (*Chanos chanos*), each weighing 300–450 grams and measuring 25–28 cm in length, were sourced from a local market in Muñoz, Nueva Ecija, Philippines. The market's production was intended for local consumption, including supply to local restaurants. The fish were transported to the laboratory and placed in holding tanks before preparation.

To ensure humane handling, the fish were first stunned using a solution containing 25 kg of table salt per kilogram of fish. They were then gently placed in a water bath (2–4°C) inside a separate container filled with ice to prevent direct contact. The fish remained in the cool water bath for at least 10 minutes before removal. Pithing was performed to ensure humane treatment. The fish were then gutted, washed, and prepared for further procedures.

Control samples were commercially sourced from a cooperative in Bataan, Philippines. Each milkfish weighed

350–450 grams and was smoked using hardwood sawdust. Control samples were selected based on the most recent production date or aligned with the production of smoked milkfish using the three smoking materials

The Smoking Process

The hot smoking process was applied uniformly across all smoking materials to minimize variability caused by process differences. A total of 54 cleaned milkfish samples were used, with 18 fish assigned to each smoking treatment.

Each batch of 18 fish was processed separately. The samples were first brined in a solution containing 2,500 mL of water, 250 mL of vinegar, 250 grams of salt, and 8 grams of additives (ginger and garlic) for every 5 kg of fish. They were soaked for 60 to 120 minutes to enhance flavor absorption.

After brining, each batch was individually blanched in a boiling saltwater solution (5,000 mL of water and 500 grams of salt) at 80°C for 10 minutes. The fish were then pre-dried in the shade for 30 to 60 minutes to enhance surface gloss, remove excess moisture, and prevent case-hardening. Before smoking, the smoking chamber and trays were thoroughly cleaned to prevent microbial contamination.

For smoking, the fish racks were placed inside the traditional smokehouse, where the temperature was monitored and maintained at 80 ± 2°C for two hours, regulated by adjusting the smoking materials. The temperature inside the smoking chamber was monitored using a calibrated thermometer. The fish were turned every hour to ensure even cooking and consistent smoke absorption.

Humidity within the smoking chamber was indirectly regulated by adjusting the airflow (vent opening) and the rate of wood combustion. The generation of smoldering smoke was maintained to prevent excessive drying or dripping of the fish.

After smoking, the samples were removed, transferred to a clean tray for cooling, labeled, and sent for evaluation.

PAH and Microbial Load Analysis

The samples were sent to an accredited laboratory for analysis immediately after production to ensure accurate assessment. The samples were analyzed at a laboratory accredited by the Philippine Accreditation Bureau (PAB) for ISO/IEC 17025 standard procedures and certified by the Food and Drug Administration (FDA).

The study used a completely randomized design with treatments replicated three times, where each replicate consisted of six fish. A random sampling method, generated using Microsoft Office Excel, was used to ensure unbiased selection and accurate representation of the overall quality of each treatment. A total of 9 samples per treatment (SWS, MWS, and CCS) were selected for analysis. Each of these fish was used for both PAH and microbial load analysis.

The PAH content of the smoked fish samples was determined using a Shimadzu GCMS-QP2010 Series Gas Chromatography–Mass Spectrometry (GC–MS) system (Shimadzu Corporation, Japan), following standard chromatographic procedures for PAH analysis. Microbial load was assessed using the Standard Plate Count method, adhering to ISO/IEC 17025 accredited laboratory procedures. Specifically, Plate Count Agar was used as the culture medium (El-Gendy *et al.*, 2024; Saelens & Houf, 2022). Plates were incubated at 30°C for 48 hours to determine the mesophilic aerobic count, consistent with general guidelines from ISO 4833-1:2013 (El-Gendy *et al.*, 2024). Results were expressed as Colony Forming Units per gram (CFU/g) (El-Gendy *et al.*, 2024).

Only samples with microbial counts within the acceptable limit of 5×10^5 CFU/g, as specified by the Food and Drug Administration (Food and Drug Administration, 2022), and PAH4 levels within the maximum allowable limit of 30 µg/kg set by the European Food Safety Authority (Afé *et al.*, 2021; Catena *et al.*, 2020; Racoviţă *et al.*, 2021), were subjected to sensory evaluation. Samples exceeding these limits were properly disposed of following food safety and waste management protocols.

The turnaround time for laboratory analysis was seven days. During this period, all labeled samples were stored in clean, airtight containers at 4°C to minimize microbial growth and maintain quality.

Sensory Evaluation

Sensory evaluation was conducted using a 9-point hedonic scale (Nicolás *et al.*, 2010; Wichchukit & O'Mahony, 2015), a widely accepted method for measuring consumer acceptability (Lim & Fujimaru, 2010; Wichchukit & O'Mahony, 2015), where 9 represented 'like extremely' and 1 represented 'dislike extremely'. The University staff and student participants were selected based on their self-reported frequency of smoked fish consumption, with preference given to those who consumed fish at least once per week.

The samples, smoked using SWS, MWS, CCS, and control, were assessed based on color, aroma, taste, and overall acceptability. Treatments (SWS, MWS, CCS, Control) were done in triplicate using a Completely Randomized Design.

The evaluation was conducted in a well-lit, temperature- and humidity-controlled room to minimize environmental factors that could affect sensory perceptions. The room was odor-neutral and well-ventilated to eliminate lingering smells that might influence the panelists' judgments. Standardized lighting ensured uniform color perception across all samples.

Each panelist was seated individually to reduce distractions and prevent influence from others' reactions. The samples, weighing approximately 15 grams each and placed on a small aluminum paper plate, were coded with three-digit numbers to ensure blind evaluation. Samples were prepared for each of the four treatment groups (three smoking materials and a control group), with duplicates available in case panelists needed to repeat an evaluation. The sensory evaluation was conducted in a single session, with samples presented in a random order to each panelist, generated using Microsoft Office Excel. Panelists were provided with water and unsalted crackers to cleanse their palates between tastings, ensuring that each sample was evaluated independently.

Statistical Analysis

Descriptive statistics were used to analyze the polycyclic aromatic hydrocarbon content and microbial load of smoked milkfish samples, assessing variations in safety and quality. Microsoft Office Excel was used to create tables and charts for data presentation.

For sensory analysis, the normality of the data was assessed using the Shapiro-Wilk test. As the sensory data did not meet the assumption of normal distribution, the non-parametric Kruskal-Wallis H-test was applied. Results were considered statistically significant at $p < 0.05$ and statistically insignificant at $p > 0.05$.

Results and Discussion

PAH Concentration

Table 1. Polycyclic aromatic hydrocarbons (PAHs) levels in milkfish smoked using different smoking materials

Smoking Material	PAH Levels (µg/kg)	LOD (µg/kg)	EU Regulatory Limit (µg/kg)
[1]SWS	< 10[a]	10	30
[2]MWS	< 10[a]	10	30
[3]CCS	< 10[a]	10	30
[4]Control	< 10[a]	10	30

[a] <10 = not detected, below the limit of detection (10 (µg/kg); [1] Santol Wood shavings; [2] Mango Wood shavings; [3] coconut shells ;
[4]hardwood sawdust

The analysis of PAH concentrations in smoked milkfish across all smoking materials—santol wood shavings, mango wood shavings, and coconut shellsshowed levels below the detection limit of the gas chromatography-mass spectrometry method used (< 10 µg/kg), as confirmed by an accredited laboratory (Table 1). This indicates that none of the smoking materials contributed detectable PAH levels above this threshold, including key carcinogenic compounds such as benzo[a]pyrene (BaP), benz[a]anthracene, benzo (BaA), fluoranthene (BbF), and chrysene (CHR) (Aksun Tümerkan, 2022).

This finding suggests that the smoking conditions employed in this study were effective in minimizing the formation and deposition of PAHs, thereby ensuring compliance with stringent safety standards. The European Commission (EC Regulation No. 835/2011) sets maximum limits for PAH4 at 30 µg/kg in smoked fish products (Afé *et al.*, 2021; Catena *et al.*, 2020; Racoviţă *et al.*, 2021). Some regulations have even stricter limits, with the EC setting a maximum limit of 12 µg/kg for PAH4 (the sum of BaP, CHR, BaA, and BbF) in smoked fish (Aksun Tümerkan, 2022). Given these benchmarks, the reported <10 µg/kg PAH4 for all samples indicates that levels were well below established regulatory limits.

While previous studies, such as the work by Whenu *et al.* (2022), have reported elevated PAH levels in smoked *Clarias gariepinus* using *Mangifera spp.* (mango wood), suggesting that variations in smoking parameters and raw materials affect PAH accumulation. The present study's results contrast with such findings. The absence of detectable PAHs at the method's sensitivity suggests effective control of smoking parameters during the process. This low PAH presence could be attributed to the controlled hot smoking temperature maintained at 80 ± 2 °C and the specific design of the traditional smokehouse used. Controlled combustion and smoke generation, as implemented in this study, are known to produce a smoke profile and reduce PAH deposition compared to direct smoking processes where food is exposed directly to flames or poorly controlled high temperatures (Afé *et al.*, 2021; Catena *et al.*, 2020; Nizio *et al.*, 2023). Factors such as wood composition, smoking techniques, and environmental conditions significantly influence PAH formation in smoked products (Nizio *et al.*, 2023; Racoviţă *et al.*, 2021). The preparation steps, such as bark removal from the wood, further contributed to reducing potential sources of unwanted compounds (Nizio *et al.*, 2023).

Microbial Load

Table 2. Mean microbial load of smoked milkfish processed using different smoking materials

Smoking Material	Mean Microbial Load (CFU/g) (TPC)	PNS Acceptable Limit (CFU/g)
[1]SWS	>6,500	5 x 10^5
[2]MWS	>6,500	5 x 10^5
[3]CCS	370	5 x 10^5
[4]Control	>6,500	5 x 10^5

[1]Santol Wood shavings; [2]Mango Wood shavings; [3]coconut shells; [4]hardwood sawdust

The microbial load, measured as the standard plate count, varied among smoking materials, as shown in Table 2. Fish smoked with coconut shells had the lowest mean microbial count at 370 CFU/g, while those smoked with santol wood shavings and mango wood shavings generally showed higher counts, with some exceeding 6,500 CFU/g. However, all samples, irrespective of the smoking material, remained well below the maximum allowable limit of 5 x 10^5 CFU/g as set by the Food and Drug Administration (FDA) (Food and Drug Administration, 2022) for smoked fish.

The significantly lower microbial load observed in fish smoked with CCS may be attributed to the antimicrobial compounds naturally present in coconut shell smoke. Coconut shell pyrolysis is known to yield smoke rich in phenolic compounds (e.g., guaiacol, cresols, catechol), organic acids (e.g., acetic acid), and carbonyls (Malaka *et al.*, 2021; Hasibuan *et al.*, 2024). These phenolic compounds are highly effective in disrupting bacterial cell membranes, denaturing proteins, and inhibiting enzyme activity, thereby impeding microbial growth (Chen *et al.*, 2022). Carbonyl compounds can penetrate cell walls and inactivate intracellular enzymes, while organic acids contribute to a lower pH environment, which is unfavorable for many spoilage microorganisms (Ekonomou *et al.*, 2020).

In contrast, the higher microbial load in SWS and MWS smoked fish, while still within safe limits, suggests that the smoke chemistry or smoking efficiency, such as penetration, concentration of active compounds from these materials, may offer comparatively less microbial inhibition under the conditions tested. The pre-smoking treatments, such as brining and blanching, along with the controlled hot smoking temperature of 80 ± 2 °C, collectively contributed to the overall reduction of microbial populations across all samples, as heat and smoke components have a synergistic preservative effect (Elazzazy & Abdallah, 2023; Barros *et al.*, 2023; Ekonomou *et al.*, 2020). Brining and blanching steps before smoking are known to significantly reduce initial microbial loads on fish surfaces (Abel *et al.*, 2022; Barros *et al.*, 2023).

Sensory Evaluation of Smoked Milkfish

a. Individual Sensory Attributes

Table 3. Mean liking scores for individual sensory attributes of smoked fish samples (N=85) (Stone, 2018; De Kock & Magano, 2020).

Sample Identity	Color	Aroma	Taste	Mouthfeel
[1]Control	176.37	174.44	174.15	181.84
[2]SWS	167.85	170.09	170.66	158.8
[3]CCS	172.29	160.68	149.86	158.18
[4]MWS	165.48	176.79	187.33	183.18
Chi-Square	0.6709	1.4384	6.9134	5.5054
DF	3	3	3	3
Pr>Chi-Square	0.88	0.6966	0.0747	0.1383

[1]Hardwood sawdust [2]Santol Wood shavings [3]Coconut shells [4]Mango Wood Shavings

No statistically significant differences were found in individual sensory attributes (p > 0.05), as shown in Table 3. However, MWS-smoked samples generally showed numerically higher mean ranks for aroma (176.79), taste (187.33), and mouthfeel (183.18) compared to other treatments. While these differences were not statistically significant, they suggest a numerical trend or tendency towards preference for MWS-smoked fish in these attributes, indicating that variations in smoking materials had a minimal, but perceptible, impact on sensory perception.

Previous studies, such as Swastawati *et al.* (2012) and Leksono & Ikshan (2020), have reported that smoking materials can influence the sensory attributes of smoked fish. However, the extent of these effects depends on various factors, including smoking duration, temperature control, and the specific chemical composition of the wood smoke. The lack of statistically significant differences in individual attributes might suggest that the overall controlled smoking process (brining, blanching, consistent temperature) homogenizes some sensory impacts, or that the differences imparted by the woods were subtle to the consumer panel.

b. Just About Right Analysis

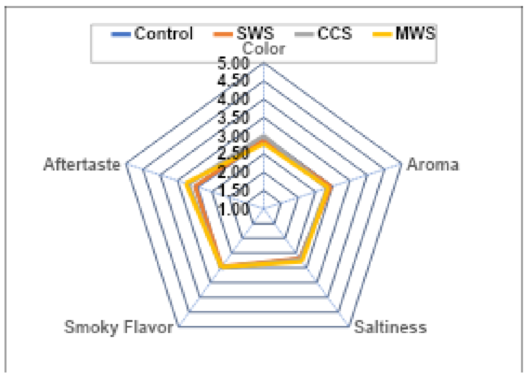

Figure 3. Spider chart for Just about right sensory mean scale. The scale ranges from 1 = much too low, 2 = a little too low, 3 = just about right, 4 = a little too much, and 5 = much too much) (Assogba *et al.*, 2024).

The Just About Right analysis, visually represented as a spider chart (Figure 3), presents the mean sensory liking scores of the smoked milkfish samples across five attributes: color, aroma, saltiness, smoky flavor, and aftertaste. A value of 3 indicates the ideal intensity of each sensory attribute. The dashed reference circle at value 3 represents the optimal sensory balance (Gere *et al.*, 2015). Treatments with mean values closest to this reference line are considered well-balanced in terms of five attributes. Samples with values above 3 exhibit stronger attribute intensity, while values below 3 indicate weaker perception (Pariès *et al.*, 2022). Based on the chart, MWS scores appear to be closer to the reference line across most attributes, particularly for color, saltiness, and smoky flavor, as its line extends further on these axes compared to the other samples. SWS and CCS show relatively similar profiles, often scoring higher than the Control, especially in attributes like Saltiness and aftertaste. Control generally exhibits lower scores across most attributes, compared to the other samples.

c. Overall Acceptability

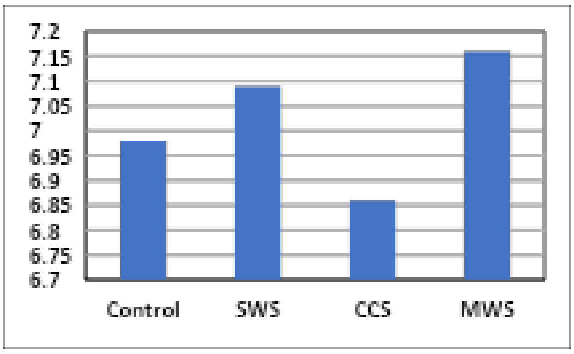

Figure 4. Mean sensory scores of smoked milkfish on a 9-point hedonic scale, evaluated by 85 respondents. The scale ranges from 1 = Dislike Extremely, 2 = Dislike Very Much, 3 = Dislike Moderately, 4 = Dislike Slightly, 5 = Neither Like nor Dislike, 6 = Like Slightly, 7 =

The Kruskal–Wallis test revealed no statistically significant differences in the overall acceptability of the smoked milkfish samples among the four treatments at the 5% significance level (p = 0.39). Although the MWS-smoked samples obtained the highest mean rank for overall acceptability (181.98), this difference was not statistically significant, indicating that the products were similarly acceptable to the panelists. This nuanced flavor profile from mango wood may have been perceived as more appealing or balanced by the panelists, leading to the numerically higher mean scores for aroma, taste, and mouthfeel, and ultimately contributing to its higher acceptability, though not statistically significant. The control, smoked with generic hardwood sawdust, might present a more traditional, stronger, or harsher, smoky flavor, although familiar was less preferred than the MWS by some panelists.

Notably, the MWS treatment received a mean hedonic score of 7.2 ("Like Moderately"), suggesting a slight preference trend; however, all treatments were generally well-accepted, as illustrated in Figure 4.

d. Pairwise Comparisons

Table 4. Result of the Dunn's test

Group 1	Group 2	R-Mean	P-Value
Control	SWS	5.311765	0.698696
Control	CCS	27.52353	0.044887
Control	MWS	38.03529	0.005576
SWS	CCS	22.21176	0.105527
SWS	MWS	32.72353	0.017096
CCS	MWS	10.51176	0.443665

Pairwise comparisons using Dunn's test (summarized in Table 4) revealed statistically significant differences in overall acceptability between certain treatments. Specifically, a significant difference was observed between the Control and MWS (p = 0.0056), control and CCS (p= 0.044887), and between SWS and MWS (p = 0.0171). These findings suggest that mango wood shavings significantly impacted the sensory attributes, leading to a different overall acceptability perception compared to both the commercial control and santol wood shavings. However, the test showed no significant differences between SWS and CCS (p = 0.105527), and between CCS and MWS (p = 0.443665), implying these pairs might impart more similar sensory experiences in some attributes.

The observed numerical preference for MWS-smoked milkfish and the statistically significant differences in pairwise comparisons for overall acceptability involving MWS can be attributed to the unique volatile compounds released during the pyrolysis of mango wood. Different wood types produce varying compositions of phenolic compounds, carbonyls, and organic acids, which are fundamental to flavor development in smoked foods (Hasibuan *et al.*, 2024; Savin *et al.*, 2024). While specific detailed chemical analyses of mango wood smoke for food processing are less common, fruit woods generally tend to produce a milder, sweeter smoke compared to more robust hardwood smokes, potentially with fruity or floral undertones that could be imparted by specific esters or terpenes in the wood (Feng *et al.*, 2022).

The lack of statistically significant differences between SWS and CCS, and between CCS and MWS in some attributes, suggests an overlapping chemical composition or intensity of smoke components, leading to similar sensory perceptions (Savin *et al.*, 2024). For instance, coconut shells produce a smoke rich in phenolics and acids, which impart distinct smoky and sometimes slightly acidic notes (Hasibuan *et al.*, 2024). If SWS and MWS also produce certain flavor-active compounds that contribute to a generally pleasant, however different, profile, this could explain the lack of wider statistical discrimination among them. The blend of acids, alcohols, carbonyls, lactones, and phenols in wood smoke critically shapes the organoleptic quality of smoked products (Savin *et al.*, 2024).

Conclusion

This study evaluated the effects of three different locally available smoking materials—mango wood shavings, santol wood shavings, and coconut shells—on the sensory properties, polycyclic aromatic hydrocarbon content, and microbial load of smoked milkfish.

The analysis of PAH levels confirmed that all smoked fish samples had PAH concentrations below the detection limit of 10 μg/kg for smoked fish. This demonstrates the safety of the smoking processes employed.

Microbial analysis showed that all samples remained well within microbiological safety limits, as set by the Food and Drug Administration (5 x10^5 CFU/g). Among the materials tested, CCS-smoked fish exhibited the lowest microbial load (370 CFU/g), suggesting its superior antimicrobial efficacy.

Sensory evaluation results, while showing no statistically significant differences in overall acceptability via the Kruskal-Wallis test, indicated that all three smoking materials produced smoked milkfish with comparable and generally moderate acceptability. Pairwise comparisons, however, revealed a significant difference in overall acceptability when MWS was compared to control and SWS. Furthermore, the Just-About-Right analysis highlighted that MWS samples offered the most balanced sensory profiles, aligning closely with ideal intensities for most attributes. While CCS and SWS showed potential for specific attributes,

This study makes a significant contribution by providing the first comparative evaluation of these specific locally available tropical woods (santol, mango, and coconut) for smoking milkfish. It explicitly demonstrates their ability to achieve safe PAH thresholds under traditional smoking conditions prevalent in the region, thereby filling a critical gap in scientific literature for these materials in the context of regional food processing.

Further research is warranted to optimize smoking parameters (e.g., precise temperature control, smoking duration, and smoke density) for each of these local wood types. This optimization should aim to further enhance microbial safety, fine-tune specific sensory attributes to potentially increase their balance in JAR assessments, and confirm consistent low PAH levels. Investigating the precise chemical profiles of the smoke generated from SWS and MWS would provide insights into their sensory contributions.

Finally, the important findings regarding the safety and acceptability of these local smoking materials should be effectively disseminated to local processors, farmers, and agripreneurs involved in smoked fish production. Training programs on best practices for smoking, hygiene, and quality control can facilitate the adoption of these effective methods, empower local communities, and enhance food safety standards within the industry. This will allow for capitalization on the unique sensory profiles offered by local wood types while ensuring compliance with national and international food safety regulations.

Ethical Statement

All procedures were reviewed and approved by the ethics committee of CLSU. Informed consent was obtained from all participants, who were assured of confidentiality and voluntary participation. The study adhered to the Declaration and relevant ethical guidelines.

Conflict of Interest Statement

The authors declare no conflicts of interest concerning the conduct or publication of this research. All procedures were carried out in compliance with institutional and ethical guidelines, and no financial or personal relationships existed that could have affected the study's outcome.

Acknowledgements

The authors would like to acknowledge the Department of Agricultural and Biosystems Engineering, College of Engineering, and the Department of Food Science and Technology of Central Luzon State University, as well as the Department of Science and Technology – Science Education Institute (DOST-SEI) through the CLSU-ERDT Program, for serving as the funding source for this study.

Declaration of Generative AI and AI-Assisted Technologies

During the preparation of this work, the author(s) utilized Grammarly only for grammar checking and language clarity. Following the use of this tool/service, the author(s) conducted a review and made necessary modifications, assuming full responsibility for the content of the publication.

Data Availability

All data supporting the findings of this study are available within the paper.

Author Contributions

DO: conceptualization, investigation, data curation, writing an original draft, reviewing and editing; EV: conceptualization, supervision, data curation, writing-review and editing; **WM:** conceptualization, reviewing and editing; **TS:** conceptualization, reviewing and editing; **SB:** conceptualization, reviewing and editing. All authors have read and agreed to the published version of the manuscript.

Funding

This study was supported by the DOST-SEI through CLSU-ERDT. No specific grant number is associated with this support.

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