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## **RESEARCH ARTICLE**

### PRODUCTIVITY EVALUATION OF THREE OYSTER MUSHROOM SPECIES (*PLEUROTUS CORNUCOPIAE*, *PLEUROTUS CITRINOPILEATUS*, *PLEUROTUS OSTREATUS*) ON VARIOUS ORGANIC SUBSTRATES (CÔTE D'IVOIRE)

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ARTICLE INFO	ABSTRACT
Article History: Received 28 <sup>th</sup> November, 2024 Received in revised form 04 <sup>th</sup> December, 2024 Accepted 18 <sup>th</sup> December, 2024 Published online 30 <sup>th</sup> January, 2025	<ul> <li>Wild mushrooms are vitally important non-timber forest products. However, changes in biodiversity and the effects of climate change are influencing their seasonal availability. Different types of substrate were formulated for this study, including three based on sawdust and one based on banana leaves. Before spawning, the culture bags were sterilised and then divided into three groups according to the type of oyster mushroom species (<i>Pleurotus cornucopiae, Pleurotus ostreatus and Pleurotus citrinopileatus</i>). After spawning, the culture bags were stored in a dark room until they were completely invaded. They were then stored in a fruiting room. The parameters assessed were: stipe height, number of clumps, number of runts,</li> </ul>
Keywords:	average number of plants per clump, cap diameter, carpophore mass and yield. The results of the statistical
Productivity, Organic Substrates, Mushroom.	analyses showed that the substrates had an influence on changes in the various parameters considered. The study carried out on the evaluation of the production potential of various oyster mushroom species showed that <i>Pleurotus cornucopiae</i> was the species that recorded a high yield on sawdust-based substrates compared with the other oyster mushroom species.

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## **INTRODUCTION**

Tropical ecosystems are rich in natural resources, including nontimber forest products (Loubelo, 2012). Among these resources are mushrooms (Pitta et al., 2020), which constitute a distinct organic kingdom with their own classification. Mushrooms have been used for centuries in Asia due to their traditionally recognized health benefits. However, their use extends beyond health, as some mushrooms are edible (Cassar, 2016). In Africa, mushrooms are significant non-timber forest products with nutritional and economic value (Ndoye et al., 2007). They are utilized by local populations for their well-being (Boot, 1997; Dossou et al., 2012) and serve as an important nutritional resource during difficult periods (Härkönen et al., 2003; EyiNdong et al., 2014; Degreef et al., 2016). Additionally, they provide a source of income for households (De Kesel et al., 2002).In Africa, the experimental and/or industrial cultivation of edible mushrooms has mainly been limited to strains imported from Europe or Asia. Various factors suggest that oyster mushroom cultivation could be promising in Africa. The thermal requirements for oyster mushroom fruiting align relatively well with the average, minimum, or maximum temperatures of different African regions.

However, in recent years, climate change has impacted the seasonal availability of fruiting bodies (De Kesel *et al.*, 2002; Mpulusu *et al.*, 2010), leading to a decline in mushrooms and affecting harvests and sales in local markets worldwide (Oei, 1993; Nieuwenhuijzen, 2007). Cultivating mushrooms emerges as an alternative to address their availability and serves as a profitable activity for African farmers. This study evaluated the productivity of three oyster mushroom species (*Pleurotus cornucopiae, Pleurotus citrinopileatus, Pleurotus ostreatus*) on various organic substrates to improve mushroom production in Côte d'Ivoire. The objectives were to determine the effects of mushroom species, fruiting substrates, and their interactions on production parameters and fruiting body yields.

### **METHODOLOGY**

**Study Area:** The study was conducted in Courandjourou (6°13'47" N, 5°6'57" W), a village in the Taabo locality (Figure 1), which serves as the departmental capital of the Agnéby-Tiassa region. This area features a transitional tropical climate influenced by the Baoulé and Attiéen climates, characterized by four seasons with monthly rainfall ranging from 140 to 190 mm. The annual average temperature and humidity are 32.18°C and 78%, respectively. The region is part of the mesophilic sector of the Bandama basin, featuring semi-deciduous vegetation and ferrallitic soils interspersed with eutrophic tropical brown soils derived from basic rocks.

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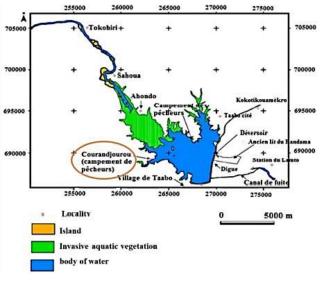


Figure 1. Map of the study area

**Biological Material:** Three strains of edible mushrooms were used in this study:

- *Pleurotus ostreatus*: A locally cultivated strain obtained from a mushroom grower in Bouaké.
- *Pleurotus cornucopiae* and *Pleurotus citrinopileatus*: Imported strains from France.

**Collection and Processing of Agricultural and Forestry Waste:** Agricultural and forestry waste used in this study was collected separately in Taabo. Dried banana leaves were harvested on-site, chopped into 1–5 cm pieces, and treated for use. Sawdust and rice bran (as an additive) were collected from a sawmill and a mill, respectively, in 100 kg bags. Sawdust-based substrates underwent aerobic composting for 40 days with regular turning. Dried banana leaves were prepared following modified methods by Pitta *et al.* (2020) and Kiyuku *et al.* (2008).

Substrate Sterilization and Bag Preparation: Once the substrates had been prepared, the polypropylene, heat-resistant sachets (33cm x 12cm) were filled with additives before being sterilised. Sterilisation was carried out using steam in a 200 L drum. A wooden tripod support 20cm high was placed at the bottom of the barrel and filled with water up to the height of the support. The closed substrates were placed vertically on the support and stacked on top of each other until the barrel was full. Sterilisation lasted 3 hours after the first vapours appeared. Once sterilisation was complete, the culture bags were cooled completely and then divided into three groups according to the type of oyster mushroom species used for the study and for each substrate. Before any handling, it was essential to disinfect the hands and the equipment to be used. Inoculation was carried out under aseptic conditions in an 'inoculation' room very early in the morning, away from direct sunlight. About two tablespoons of spawn were applied to the front of the substrate, taking care to reseal the culture bags.

**Substrate Formulation:** In this study, four (04) compost formulations were developed from the waste collected, including three based on sawdust and one based on banana leaves. For this study, the standard formulation (Sa) considered was that used by Ivorian myciculturists. This was taken as the control formulation. Thus, we have the following substrates:

- Substrate Sa (control): 70% sawdust + 10% rice bran + CaCo31%,
- Substrate Sb: 85% sawdust +13.5% rice bran + 1.5% CaCo3,
- Substrate Sc: 90% sawdust + 8.5% rice bran + CaCo3 1.5
- Substrate Fb: Banana leaves + 10% rice bran + CaCo31%.

*Experimental Design:* The experimental set-up used in this study is a split plot set-up of three blocks constituting the three species, in which four microplots corresponding to the different treatments are distributed. Each treatment included 15 culture bags spaced 1 cm apart.

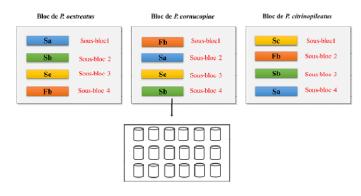


Figure 3. Expérimental setup

#### Data Collection and Analysis

**Production parameters:** Each week during the fruiting phase, various parameters were recorded in bags according to substrate and species. These were stipe height (cm), carpophore diameter (cap, cm), number of clumps, number of runts, average number of plants per clump and carpophore mass (g).

- The height of the stipe was obtained using a graduated ruler. The measurement was taken from the base of the stipe at the point of attachment to the substrate to the top (at the point of insertion into the cap);
- The diameter of the carpophore was measured using a scale, taking the length of the carpophore in both directions. The average was then calculated using the following formula: (L1 + L2) / 2.
- The number of clumps, the number of runts (unevolved primordia) and the average number of plants per clump were obtained by manual counting.
- The mass of the carpophores was obtained using an electronic balance after the mature carpophores had been harvested. This was generally done 3-4 days after the primordias had formed.

*Crop yield:* The yield expressed as a percentage was determined at the end of the experiment on each culture substrate according to the following mathematical formula:

r = (mass of fresh carpophores (kg)/mass of substrates (kg)) \* 100

The data collected was entered using EXCEL 2013. Analyses of variance (Anova) and comparisons of means using the FISHER LSD test were performed using STATISTICA version 7.1 software. Differences were considered significant at the 5% threshold (means followed by different letters). This difference is affirmed when the probability (P-value) obtained for one factor or for the combination of the two factors is less than 5% (our significance threshold).

### RESULTS

# Effects of culture substrates on oyster mushroom production parameters

Stipe height: Statistical analysis of the stipe height data for the three oyster mushroom species grown on different substrates indicates that the fruiting substrates considered significantly influenced the heights (P<0.05). The Sa substrate induced the greatest height of the stipes of *Pleurotus ostreatus* (PL) compared with PJ, whose heights were improved by the Sc substrate (3.90 cm). PG saw the height of its

stipes influenced by the substrates Sa and Sc, the maximum being recorded with Sa (5.16 cm). However, substrates Sa, Sb and Fb had the same influence on the height development of *Pleurotus citrinopileatus*.

were influenced by the Sa and Sc substrates belonging to the same homogeneous group, the maximum being recorded with Sa (5.16 cm). The banana leaf substrate obtained the lowest value for the number of clumps  $(0.50\pm0.26)$ .

#### Table 1. Influence of fruiting substrates on the evolution of the number of oyster mushroom clumps

	Fruiting Substrates								
Species	Sa	Sb	Sc	Fb	P-value	F			
PG	5,16±0,22b	2,96±1,99a	4,76±0,79b	2,43±1,21a	0,006	5,809			
PJ	1,05±0,83a	1,71±1,71a	3,90±0,61b	1,88±1,17a	0,008	5,607			
PL	5,19±0,60c	2,95±0,69b	0,00±0,00a	0,00±0,00a	0,000	148,998			

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

#### Table 2. Influence of fruiting body substrates on the development of the number of oyster mushroom clumps

	Fruiting Substrates								
Species	Sa	Sb	Sc	Fb	P-value	F			
PG	2,56±0,68c	1,53±0,84b	2,40±0,36c	0,50±0,26a	0,000	13,006			
PJ	0,70±1,56a	0,73±0,72a	1,50±0,42a	1,00±0,57a	0,520a	0,784			
PL	0	0	0	0	0	0			

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

#### Table 3. Influence of fruiting body substrates on the evolution of oyster mushroom numbers

Fruiting Substrates								
Species	Sa	Sb	Sc	Fb	P-value	F		
PG	6,33±1,56bc	4,60±2,45ab	7,39±1,12c	2,76±1,56a	0,003	6,761		
PJ	1,06±2,38a	3,53±3,82ab	5,50±1,79b	1,60±1,14a	0,048	3,282		
PL	15,36±3,33c	4,93±1,78b	0,00±0,00a	0,00±0,00a	0,000	73,30		
1								

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

#### Table 4. Influence of fruiting body substrates on the development of oyster mushroom cap diameters

Fruiting Substrates								
Sa	Sb	Sc	Fb	P-value	F			
5,15±1,23b	2,98±1,99a	4,62±1,29ab	2,68±1,18a	0,042	3,416			
0,63±1,41a	1,92±2,12a	4,69±0,63b	2,41±1,70a	0,006	5,821			
5,09±0,56c	2,88±0,86b	0,00±0,00a	0,00±0,00a	0,000	115,504			
	5,15±1,23b 0,63±1,41a	Sa         Sb           5,15±1,23b         2,98±1,99a           0,63±1,41a         1,92±2,12a	Sa         Sb         Sc           5,15±1,23b         2,98±1,99a         4,62±1,29ab           0,63±1,41a         1,92±2,12a         4,69±0,63b	Sa         Sb         Sc         Fb           5,15±1,23b         2,98±1,99a         4,62±1,29ab         2,68±1,18a           0,63±1,41a         1,92±2,12a         4,69±0,63b         2,41±1,70a	Sa         Sb         Sc         Fb         P-value           5,15±1,23b         2,98±1,99a         4,62±1,29ab         2,68±1,18a         0,042           0,63±1,41a         1,92±2,12a         4,69±0,63b         2,41±1,70a         0,006			

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

#### Table 5. Influence of fruiting body substrates on changes in oyster mushroom mass

Fruiting Substrates								
Species	Sa	Sb	Sc	Fb	P-value	F		
PG	37,40±6,47b	19,50±18,31a	43,63±11,22b	15,10±9,95a	0,005	6,287		
PJ	3,60±8,04a	11,96±14,81a	46,70±5,90b	15,26±8,73a	0,000	17,990		
PL	31,40±10,33c	17,36±4,80b	0,00±0,00a	0,00±0,00a	0,000	35,577		

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

#### Table 6. Influence of Fruiting Substrates on the Yield of Oyster Mushrooms

	Fruiting Substrates								
Species	Sa	Sb	Sc	Fb	P-value	F			
PG	24,93±4,31b	13,00±12,20a	29,08±7,48b	10,06±6,63a	0,005	6,287			
PJ	2,40±5,36a	7,97±9,87a	31,13±3,93b	10,17±5,82a	0,000	17,990			
PL	20,93±6,89c	11,57±3,20b	0,00±0,00a	0,00±0,00a	0,000	35,577			

Values followed by the same letter in a row are statistically identical at the 0.05 threshold. PG: *Pleurotus cornucopiae*; PJ: *Pleurotus citrinopileatus*; PL: *Pleurotus ostreatus*; Sa: 70% sawdust + 10% rice bran + 1% CaCO3; Sb: 85% sawdust + 13.5% rice bran + 1.5% CaCO3; Sc: 90% sawdust + 8.5% rice bran + 1.5% CaCO3; Fb: 10% rice bran + 1% CaCO3; F: Frequency; P-value: Probability.

*Number of clumps:* Statistical analysis of the clump number data for the different oyster mushroom species grown on the fruiting body substrates considered shows that these had a significant influence on the evolution of clump numbers (P<0.05). This influence was only visible with *Pleurotus cornucopiae* (P=0.000). The latter's parameters

Unlike PG, fruiting body substrates had no influence on the development of clump number in *Pleurotus citrinopileatus* (P=0.520). PL did not form any clumps.

Average number of plants per clump: Statistical analysis of the data on the average number of plants per clump and per culture bag of the different oyster mushroom species grown on the different substrates indicates that the fruiting body substrates considered had a significant influence on the number of plants of the carpophores (P<0.05). Substrate Sa induced the greatest number of *Pleurotus ostreatus* (PL) plants, followed by substrate Sb, whereas PJ saw its plant number improved by substrate Sc (5.50). As for PG, the average number of plants per clump was influenced by the Sc substrate, followed by the Sa substrate (Fb) obtained the lowest values, regardless of the species considered.

**Diameter of the cap:** The results of the statistical analysis of the cap diameter data for the oyster mushroom species as a function of the culture substrates indicate that the substrates had a significant influence on the diameters of the three species studied (P<0.05). The Sa substrate favoured the largest diameters of the caps of *Pleurotus ostreatus* (PL) and the Sa substrate followed by the Sc substrate improved that of *Pleurotus cornucopiae* (PG) compared with PJ, whose diameters were improved by the Sc substrate (4.69 cm). Substrates Sb and Fb belonging to the same homogeneous group had the same influence on the development of the caps of PG and PJ.

*Mass of carpophores:* The results of the statistical analysis of carpophore mass data for the various oyster mushroom species grown on different substrates show that the fruiting body substrates considered had a significant influence on mass (P<0.05). *Pleurotus ostreatus* (PL) obtained the highest average masses on the Sa substrate (31.40 g), followed by the Sb substrate (17.36 g). Unlike PL, the masses of *Pleurotus citrinopileatus* were improved by the Sc substrate (46.70 g); the Sa, Sb and Fb substrates had the same influence on the mass of the carpophores. PG saw its carpophore masses influenced by the Sa and Sc substrate (43.63 g).

*Effects of culture substrates on oyster mushroom yields:* The results of the statistical analysis on the yield of different oyster mushroom species grown on various substrates show that the fruiting substrates significantly influenced the yields (P<0.05). *Pleurotus ostreatus* (PL) achieved its highest yield on substrate Sa (20.93%), followed by substrate Sb (11.57%). In contrast to PL, the yield of *Pleurotus citrinopileatus* was improved by substrate Sc (31.13%), while substrates Sa, Sb, and Fb had the same influence on the yield of the fruiting bodies. For PG, the yield was influenced by substrates Sa and Sc, with the maximum yield obtained in cultivation bags containing substrate Sc (29.08%)

## DISCUSSION

To evaluate the production potential of different oyster mushroom species (P. cornucopiae, P. citrinopileatus, and P. ostreatus) on various organic substrates, including those based on sawdust (Sa, Sb, and Sc) and banana leaves, it was necessary to assess the influence of these substrates on the evolution of production parameters and the yield of fruiting bodies. The parameters considered were stipe height, cap diameter, cluster count, average number of stems per cluster, fruiting body mass, and fruiting body yield. The production parameters of the different oyster mushroom species were influenced by the substrates studied. For most of the production parameters considered, probabilities were less than 0.05 (P<0.05). The stipe heights of the three oyster mushroom species were generally improved by sawdust-based substrates Sa and/or Sc. The Fb substrate, formulated with banana leaves, produced moderate results but was less effective than sawdust-based substrates for Pleurotus cornucopiae and Pleurotus ostreatus. These findings differ from those of Mushagalusa et al. (2017), who found no significant influence of substrates on the parameter studied. However, they align with Lushiku (2012), who recorded lower stipe lengths with banana leaf-based substrates.Regarding cluster count, the substrates had a similar influence on this parameter for Pleurotus citrinopileatus, unlike Pleurotus cornucopiae, which saw its cluster count improve with substrates Sa and Sc, while Sb and Fb resulted in fewer abortive

clusters. Results for PJ align with Mushagalusa et al. (2017), who found no significant difference between bean leaf and banana leaf substrates on cluster count. Conversely, Pleurotus ostreatus did not form clusters during the study period but instead produced individual stipes. These results could be attributed to the inherent characteristics of each species. For parameters such as the number of stems per cluster, cap diameter, fruiting body mass, and yield, higher values were obtained with the control substrate (Sa: 70% sawdust, 10% rice bran, 1% calcium carbonate) and/or Sc (90% sawdust, 8.5% rice bran, 1.5% calcium carbonate). These formulations enhanced the evolution of these parameters for all three oyster mushroom species compared to banana leaf-based substrates, which recorded relatively low values. The total yield of substrates varied by formulation and strain, with a total yield of 16.08% on Sa, 10.85% on Sb, 20.07% on Sc, and 6.74% on Fb. These results align with Ibeghouchene&Slimani (2020), who noted that yields vary depending on substrate composition, and Chalaux (1993), Lourdes et al. (2008), and others who demonstrated that yield varies by strain. Moreover, Pitta et al. (2020) showed that sawdust-based substrates offered high fruiting body yields for the Pleurotus genus, with yields ranging from 132 to 186 g/kg of substrate. The observed results could be explained by the availability of easily assimilable nutrients. The presence of additives like rice bran in substrates increases the average mass of mushrooms per bag and positively impacts total yield. Lower yields on banana leaf-based substrates may result from low nitrogen content. According to Oei (2003), substrates with higher nitrogen content are more productive, a statement corroborated by previous studies showing greater productivity of bean leaves compared to banana leaves.

### CONCLUSION

The formulated substrates improved the productivity of all three oyster mushroom species. However, the substrate Sc (90% sawdust, 8.5% rice bran, 1.5% calcium carbonate) yielded the best results, followed by the control substrate Sa. The imported species *Pleurotus cornucopiae* and *Pleurotus citrinopileatus* adapted well to local substrates and produced good yields. Given this adaptability and productivity, the next chapter will focus on trials for producing spawn for these species.

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