



EFFECTS OF DIFFERENT SUBSTRATES AND TEMPERATURE ON THE GROWTH AND YIELD OF OYSTER MUSHROOM (*Lentinus sajor-caju* Fr.)

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Abstract

The study evaluated the effects of three saw dust substrates on the growth and yield of *Lentinus sajor-caju* at the pathology laboratory of Forestry Research Institute of Nigeria (FRIN) under indoor and outdoor temperatures of 28.6°C and 29.1°C respectively. The saw dust substrate includes; *Triplochiton scleroxylon*, *Gmelina arborea*, and *Cordia Millenii*. Wheat bran and lime (CaCO₃) were incorporated to the substrates as supplements. The mycelial growth was faster at 28.6°C giving full colonization at 3 weeks in all the substrates, while full colonization was observed in 4 weeks at 29.1°C. *T. scleroxylon* substrate recorded best heights at both indoor and outdoor temperatures with 28.1±10.2cm and 14.6±4.40 respectively. *G. arborea* substrate gave the highest yield at both outdoor and indoor temperatures with 69.5±30.6 g 53.5±10.8 g respectively. There was no significant difference on the growth and yield of *L. sajor-caju* at the two temperature ranges. Based on the results obtained, *G. arborea* saw dust was the most suitable substrate and could be recommended for the cultivation of *Lentinus sajor-caju*.

Keywords: Oyster mushroom, mycelial growth, saw dust substrates, yield, temperature

Introduction

Oyster mushroom (*Lentinus sajor-caju* Fr.) of family polyporaceae is a wild edible mushroom gaining popularity in recent years because of its high nutritional value and ability to grow on diverse agricultural wastes. Mushrooms production is considered as the second most important commercial microbial technology next to yeast (Xia *et al.*, 2016). Cultivation of wild edible mushrooms is a biotechnological process for lignocellulosic organic waste recycling (Amana *et al.*, 2013). The largest producers of mushroom is China, with 1.5 million tons, followed by the USA with 0.38 million tonnes as at 2008 with world production of about 3.4 million tons (Mishra, 2012; Kumar *et al.*, 2014). Oyster mushrooms are gaining recognition because they can be cultivated with little efforts and cost on a variety of lignocellulosic agro-wastes, with high yield and nutritional values (Gothwal *et al.*, 2012, Marlina *et al.*, 2015). Mushroom cultivation offers an eco-friendly and a cost effective strategy for effective disposal of agricultural wastes and provision of sources of food rich in protein (Bisaria *et al.*, 1987; Gothwal *et al.*, 2012; Gupta *et al.*, 2013; Marlina *et al.*, 2015). Wild species of *L. sajor-caju* contains high protein and carbohydrates, good quantities of vitamins, carotenoids

and minerals and low fat and so can be considered as nutraceutical and functional food for humans (Singdevsachan *et al.*, 2013). The interest in commercial production of wild edible mushrooms has been on the increase significantly in many parts of the world in the past few decades (Tibuhwa, 2013, Manna, 2014). Customarily, wild edible mushrooms harvesting have been obtained in the forest, especially during rain seasons, making them seasonal and not available throughout the year. The impacts of climate in addition to increase in human population, pollution, human occupational activities; such as firewood gathering, bush burning, agriculture practices and urbanization have contributed to the decline of mushrooms in the forests (Okigbo and Nwatu, 2015). The interest in cultivation of mushroom to complement the practice of harvesting from the wild is currently on the increase (Mishra, 2012). Portable logs previously colonized by mushrooms are relocated to home gardens from the wild where they are nurtured until the appearance of the next flush in some parts of West African countries (Osarenkhoe *et al.*, 2014). The process of mushroom cultivation entails the right environment and provision of appropriate media for the fungi to develop their mycelia to mycelia mass which will transform into new

fruiting bodies (Mensah, 2015). The growth and yield of Mushrooms are highly influenced by several factors such as spawn, growing media, pH, temperature, moisture content and light intensity (Kadiri and Kehinde, 1999). The high temperature (27°C - 35°C) during the day is a major constraint to farmers for mushrooms cultivation in the tropical region (Gaitán and Salmones, 2008). Furthermore, identification of suitable substrate and temperature for incubation is crucial to achieve high yield and quality mushrooms. This study evaluated the effects of indoor and outdoor temperature on the growth and yield of *Lentinus sajor-caju* on three different substrates (*Triplochiton scleroxylon*, *Gmelina arborea* and *Cordia millenii*).

Materials and Methods

Study area and sources of experimental material

The study was conducted at Pathology Laboratory of the Forestry Research Institute of Nigeria (FRIN). The Institute is situated in Jericho hill, under Ibadan North West Local Government Area of Oyo State. The area lies between latitude 7°26'N and longitude of 3°54'E. The climatic condition of the area is tropically dominated by rainfall pattern from 1400mm-1500mm annually. The average temperature is about 30°C, average relative humidity of about 80-85% (FRIN, 2018). The saw dusts were obtained at Bodija saw-mill and the wheat bran at live-stock feed mill seller both in Ibadan, Oyo State Nigeria. The spawn (Mushroom seed) were obtain at pathology laboratory of the Institute.

Preparation of media for mushroom culture

The different saw dust substrates were mixed with water and lime at rate of 1% of the saw dust and wheat bran were added at the rate of 5% base on the saw-dust dry weight. The bags were packed inside white polythene bags with mean thickness of 0.2mm containing 500g wet weight of the substrate. The openings of bags were tied with rubber band to prevent the substrate from pouring out. The sterilization was done immediately after the bagging, at the temperature of 121°C in an autoclave. After sterilization, hands, bowl and the table (to put them on) were also sterilized with methylated spirit.

Inoculation of spawn and incubation

The rubber bands were later removed before holes were punched with the aid of sterilized hole stick. Inoculation of the bags with spawn was at the rate of 5% based on

wet mass of the substrate. After inoculation of the spawn into the bags, their openings were tied back with rubber band and the bags divided into two sets: 12 bags were placed in the room, and other 12 bags placed outside in a shaded environment for incubation. The experimental design is a 3x2 factorial involving 'treatment/factor1 at 3 levels' and 'treatment/factor 2 at 2 levels' laid out in Complete Randomize Design (CRD) in four replicates.

Data collection and analysis

The daily temperature was recorded using laboratory thermometer base on sun rise and sunset (9:45am – 10:15am, 3:45pm - 4:15pm respectively). The data on mycelial growth and other parameters commenced one week after inoculation and continued weekly. The mycelial growth and the length of harvested mushroom tips (diameter of pilos) were measured weekly using a transparent meter ruler. Mycelial growth was measured in centimeters as the length of the mycelium was spreading from the mouth of the bag toward the bottom side at a weekly interval for three weeks. The harvested mushrooms were weighed on sensitive weighing scale. Data collected were subjected to Analysis of Variance (ANOVA). Treatment means were separated by Turkey's Honestly Significant Difference (THSD) at 0.05 level of significance with ASSISTAT statistical software 7.6 beta.

Results and Discussion

Effects of different substrates on mycelial running at indoor temperature (28.6°C)

Mycelial running is an extension and colonization of fungal hyphae throughout the substrate. The results of this study revealed that different substrates evaluated enhanced the mycelial running of *L. sajor-caju* at indoor temperature; however the rate of extension varied among the substrates. Spawn inoculated on *Triplochiton scleroxylon* substrate exhibited faster mycelial extension than in other substrates with 37.5cm and 73.0cm in the first and second week respectively (Fig. 1). The Mycelial extension on *Gmelina arborea* substrate was second in the order with 30.7cm and 67.7cm at the first and second week respectively. There were no significant differences on the mycelial extension among the different substrates. Full mycelia colonization was observed in all the substrates at three weeks (21 days) after spawn inoculation.

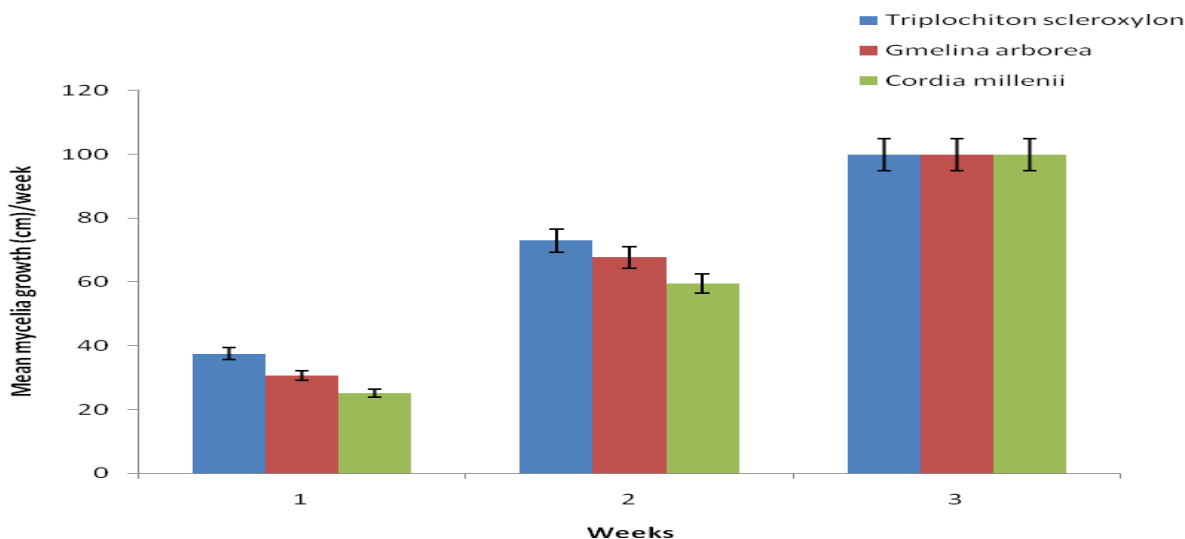


Fig. 1: Weekly mycelial growth of *Lentinus sajor-caju* on different substrates at indoor temperature
Vertical bars represent \pm S.E of means

Effects of different substrates on mycelial running at outdoor temperature (29.1°C)

Mycelial running at outdoor temperature on different substrates followed a different pattern from what was obtained at indoor temperature. *Lentinus sajor-caju* spawn inoculated on *Gmelina arborea* substrate showed faster mycelial extension with 22.5 in the first week, followed by those inoculated on *Cordia millenii* substrate with 22.2cm (Fig. 2). In the second and third

week, *Cordia millenii* substrate exhibited higher mycelial growth with 44.8cm and 69.5cm respectively. *Triplochiton scleroxylon* was the second in mycelial extension with 39.3cm and 67.0cm in the second and third week respectively. There was no significant difference on the mycelial extension among the different substrates. Full mycelial colonization on different substrates were observed at fourth week (28 days) after spawn inoculation

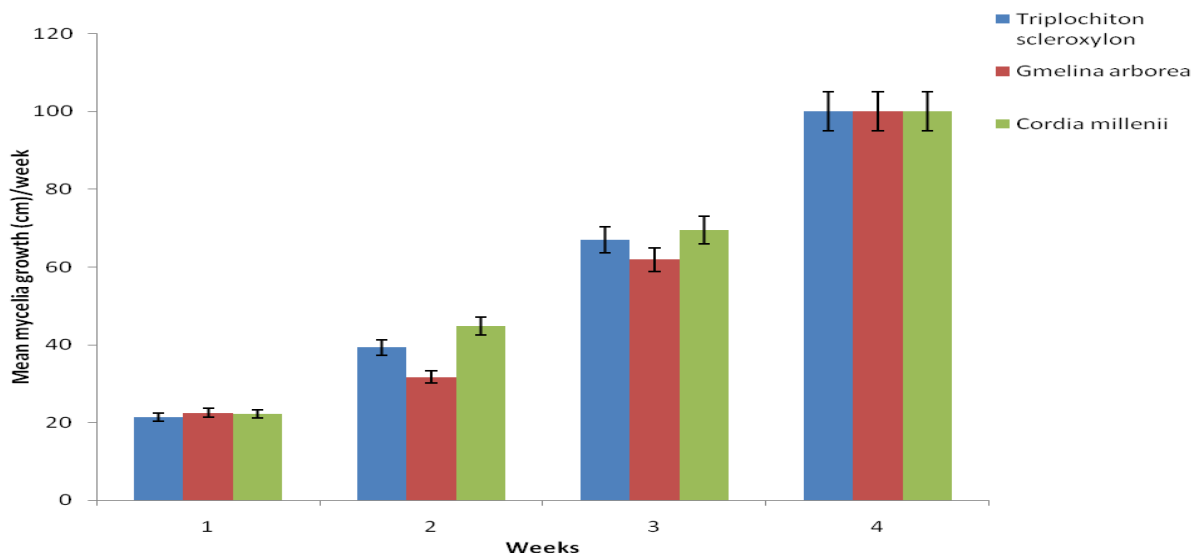


Fig.2: Weekly mycelial growth of *Lentinus. sajor-caju* on different substrates at outdoor temperature
Vertical bars represent \pm S.E of means

Effects of indoor and outdoor temperature on heights of *Lentinus sajor-caju* on different substrates

The results of this study revealed that heights of *L sajor-caju* on different substrates varied at indoor and outdoor temperature (Fig. 3). The mean indoor and outdoor temperatures during the period of the study were 28.6°C and 29.1°C respectively. The heights of the mushroom were higher at indoor temperature in all the substrates

than at outdoor temperature. Mushroom spawn inoculated on *Triplochiton scleroxylon* substrates recorded highest heights at both indoor and outdoor temperature with mean heights of 28.1 \pm 10.2cm and 14.6 \pm 4.40 cm respectively. There were significant differences ($p < 0.05$) on the heights of the mushroom grown on different substrates at indoor and outdoor temperature.

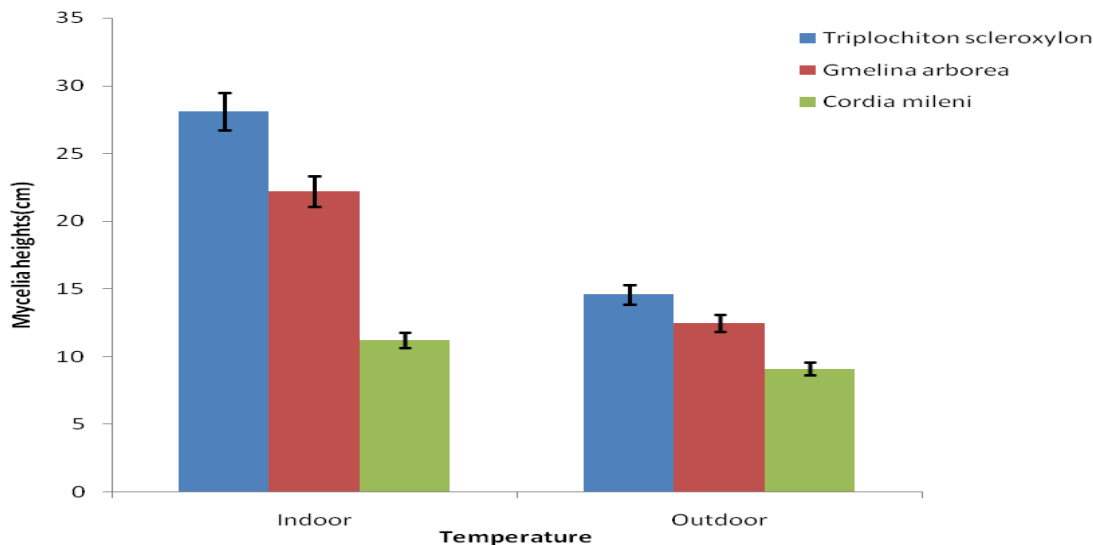


Fig. 3: The effect of outdoor and indoor temperature on the height of *Lentinus sajor-caju*
Vertical bars represent \pm S.E of means

Effects of indoor and outdoor temperatures on diameter of pilos of *Lentinus sajor-caju* on different substrates

The results of this study revealed that diameter of pilos on different substrates also varied at indoor and outdoor temperature (Table 1). The mean indoor and outdoor temperatures during the period of the study were 28.6°C and 29.1°C respectively. The diameter of the pilos was higher at indoor temperature in all the substrates

compared to outdoor temperature. Mushroom grown on *Triplochiton scleroxylon* substrates recorded highest diameter at both indoor temperature with mean values of 21.9±7.18cm, while the highest diameter was observed from *G. arborea* at outdoor temperature with 11.5±4.23cm. There were significant differences ($p < 0.05$) on the diameter of the mushroom pilos grown on different substrates at indoor and outdoor temperature

Table 1: Mean temperature effect of outdoor and indoor assay on the diameter of *Lentinus sorjo caju* .

Treatment	Diameter (cm)	
	Indoor temperature(28.6°C)	Outdoor temperature (29.1°C)
<i>Triplochiton scleroxylon</i>	21.9±7.18a	11.2±2.97a
<i>Gmelina arborea</i>	19.0±4.86b	11.5±4.23a
<i>Cordia millenii</i>	15.2±7.62c	9.17±3.43b

Means followed by the same letters within the same column are not significantly different ($p > 0.05$) using THSD

Effects of different substrates on the yield of *L sajor-caju* at the two temperature conditions

The results showed that higher yield of *L. sajor-caju* was recorded in all the substrates at outdoor temperature compared to indoor condition (Table 2). *Gmelina*

arborea substrate recorded highest yield, followed by *Triplochiton scleroxylon* with 69.5±30.6g and 61.5±23.1g respectively. *Cordia millenii* recorded almost the same yield of 42.2±22.5g and 42.0±15.1g at indoor and outdoor temperatures respectively.

Table 2: Yield of *L. sajor-caju* on different substrate at two temperature conditions

Treatment	Yield (g)	
	Indoor temperature (28.6°C)	Outdoor temperature (29.1°C)
<i>Triplochiton scleroxylon</i>	44.0±16.8b	61.5±23.1b
<i>Gmelina arborea</i>	53.5±10.8a	69.5±30.6a
<i>Cordia millenii</i>	42.2±22.5b	42.0±15.1c

Means followed by the same letters within the same column are not significantly different ($p > 0.05$) using THSD

Microorganisms, like every living organism require source of energy, optimum environmental conditions and nutrients in order to grow and reproduce (Ravimannan *et al.*, 2014). Among different saw dust substrates evaluated in this study; *Gmelina arborea* proved to be the best substrate for the mycelial growth of *L. sajor-caju*. The differences on the mycelial growth observed on different substrate may be due to

availability of different carbon sources and other required nutrients. Saw dust substrate from different tree species supplemented with wheat or rice bran and lime have been reported to enhance the growth and yield of different species of mushrooms. Debu *et al.* (2014) reported that sawdust substrate from five tree species; *Ficus carica* , *Albizia saman*, *Swietenia mahagoni*, *Leucaena leucocephala* and *Eucalyptus globules*

supplemented with 30% wheat bran and 1% lime as basal substrates enhanced the biological yield of *Pleurotus ostreatus* with highest yield obtained from *Swietenia mahagoni*. Similarly, Dey *et al.* (2006) reported that yield of oyster mushroom significantly varied with the substrates used in production. Bhuyan (2008) also reported that yield of oyster mushroom on sawdust supplemented with different levels of cow dung varied with different substrates. A recent study by Nasir1 *et al.* (2021) reported that 100% saw dust from *Vachellia nilotica* (Kikar tree) enhanced growth and yield of oyster mushroom compared to sawdust mixed with different maize residues. The mycelial running on all the substrates was faster at indoor temperature, compared to outdoor temperature in the study. Mycelial running to full colonization took about 20-21 days on all the substrates at indoor temperature, while it took average of about 27-28 days on all the substrates at outdoor temperature. These results are comparable with other similar previous studies. Ahemed (1998) reported spawn running of *P.ostreatus* was completed within 17.20 days on different substrates. Similarly, Hoa *et al.* (2015) reported that mycelia running to full colonization of *Pleurotus ostreatus* and *Pleurotus cystitidiosus* took 30- 40 days on different substrates. However, some researchers have reported shorter duration of mycelia running to full colonization of oyster mushroom. For instance, Girmay *et al.* (2016) reported that it took about 16 days for the mycelia to run on cotton seed, paper waste, wheat straw, and sawdust substrate used in their studies. Onuoha *et al.* (2009) also reported that spawn running of *Pleurotus species* took about 15 days, while Patra and Pani (1995) and Jiskani (1999) reported that spawn running of the same species took 13 to 16days. According to Chang and Miles (2004), fungal strain, growth conditions and the type substrate is a factor responsible for variation in the duration of spawn running to complete colonization. This variation could also be attributed to the differences in chemical composition of the substrates used, such as Carbon to Nitrogen ratio (Bhatti *et al.* 1987). The growth of the Mycelial in mushrooms require specific nutrients, and supplements addition can increase mushroom yield through the provision of these specific nutrients (Oei, 1996). The best mycelial growth of *L. sajor-caju* was obtained at indoor temperature mean of 28.6°C on *Triplochiton scleroxylon* substrate. This result corroborates the earlier report by Hao and Wang (2015) that the best mycelial growth of two Oyster species was obtained at optimum temperature of 28°C. Temperature is a very important environmental factor for mycelium growth of fungi. The optimum mycelial growth was obtained at outdoor temperature of 29.1°C on *Gmelina arborea*. Neelam *et al.* (2013) reported that the optimum temperature for mycelium growth of Oyster mushroom (*P. Florida*) was 25~30°C. Temperature is an important aspect in the selection of mushroom for cultivation in the tropics, where high temperatures are witnessed for nearly all the period (Hasan *et al.* 2015). The optimum growth rate for *Pleurotes species* was reported to be at 25°C (Hasan *et al.* 2015). Similarly, Zharare *et al.* (2010) reported that the maximum growth of *Pleurotus stran* was 25°C

Conclusion

Lintinus sajor-caju (Oyster mushroom) showed a promising growth potential on sawdust waste generated in the saw mill. Different saw dust substrates like *Triplochiton scleroxylon*, *Gmelina arborea* and *Cordia millenii* supplemented with wheat bran and 1% lime as basal treatment enhanced the growth and yield of *L. sajor-caju* and should be incorporated in the commercial cultivation of mushroom. Different temperature conditions influenced the growth and yield of *L. sajor-caju*. The best growth and yield of *L. sajor-caju* was obtained on *G. arborea* substrate at the mean indoor temperature of 28.6°C. Adoption of wood saw dust for mushroom cultivation would enhance profitable mushroom production, provide additional economic strategies for saw dust disposal and minimize environmental hazards associated with burning of this sawdust which is a common practice in most saw mills in developing countries.

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