BEHAVIOR OF A MEXICAN STRAIN OF *Lentinus lepideus* ON THREE SOLID MEDIA[1]

by Rigoberto Gaitán-Hernández[2], Gerardo Mata[2] and Gastón Guzmán[2]

COMPORTAMIENTO DE UNA CEPA MEXICANA DE *Lentinus lepideus* EN TRES MEDIOS SOLIDOS

RESUMEN

Se estudió el crecimiento micelial de una cepa mexicana de *Lentinus lepideus* en agar con dextrosa y papa, agar con extracto de malta y agar con dextrosa de Sabouraud, bajo cuatro diferentes temperaturas: 20,.22.5, 25 y 27.5°C. El mejor crecimiento se observó a 27.5°C en el primer medio.
PALABRAS CLAVE: *Lentinus lepideus*; crecimiento micelial; diferentes temperaturas; medios de cultivo sólidos.

ABSTRACT

Mycelial growth of a Mexican strain of *Lentinus lepideus* was studied on potato dextrose agar, malt extract agar and Sabouraud dextrose agar, at four different temperatures: 20, 22.5, 25 and 27.5°C. The best development occurred on the first medium at 27.5°C.
KEY WORDS: *Lentinus lepideus*; mycelial growth; different temperatures; solid culture media.

INTRODUCTION

Lentinus lepideus (Fr. : Fr.) Fr. is an important edible mushroom in Mexico, commonly found on *Pinus* wood (Guzmán, 1977). This fungus produces dark cubic rot which degrades the cellulose of the duramen but not degrades the lignin (Gómez-Nava *et al.*, 1969). Its tolerance to relatively high concentrations of creosote and the fact that this fungus attacks mainly the duramen, give it importance as an agent of wood rot (Dickinson, 1979; Suominen, 1973). The present study is part of a series of experiments on *L. lepideus* carried out by the authors to produce fruiting bodies on a large scale.

[1] This paper was presented at the V Congreso Nacional de Micología, at Guanajuato, in Nov. 1994.

[2] Instituto de Ecología, Apartado Postal 63, Xalapa, Veracruz 91000, México.
Received: May 4, 1995. Accepted: November 15, 1995.
Reprints' request: Rigoberto Gaitán-Hernández[2].

MATERIALS AND METHODS

The strain studied was isolated from a wild fruit body picked up at the region of Xalapa, Veracruz, growing on pine log which had been previously treated with creosote. This strain is deposited in the Strain Collection of the Instituto de Ecologia with the number IE-133, and the fruit body at XAL Herbarium.

Mycelial growth was studied on three solid culture media: potato dextrose agar (PDA), malt extract agar (MEA), and Sabouraud dextrose agar (SDA). Culture media were sterilized for 15 min at 121°C and poured in Petri dishes of 63.6 cm^2. These Petri dishes were inoculated with pieces of inoculum (8 mm diam) of the strain and incubated in the dark at 20, 22.5, 25 and 27.5°C. Five replicates were made for each treatment. Mycelial diameters were measured every third day. Color, texture, and density were observed. For color determination, the identification chart of the Royal Botanical Garden Edinburgh (1969) was used. For texture, Stalpers's terminology (1978) was used. Diameter of hyphae, clamp connections and branching type were observed by microscope with 5% KOH. The terminology of Dubovoy and Herrera (1967) was used for the description of microscopic features.

Growth data observed at different temperatures were analyzed with the BMDP (version 4V) computer program. In addition, Bonferroni's multiple comparison method was used to determine the similarity (p 0.05) of the average growth rate under each of the conditions tested.

RESULTS

Mycelium growth

Temperature 20°C. On PDA mycelium presented granular surface. After covering the whole dish in 15 days, small white primordia were formed which became peach (No. 46) colored. On MEA, after 15 days mycelium covered the whole agar and the color of the medium changed to reddish yellow (No. 12). On SDA, the color of the medium changed to mustard yellow (No. 9-H) and became brick red (No. 19). At day 12 of incubation the mean surface covered by the mycelium was 45.28 cm^2 on PDA, 44.53 cm^2 on MEA and 34.23 cm^2 on SDA (Tables 1 and 2).

Temperature 22.5°C. On PDA mycelium showed a granular surface with scant areal growth in some areas and after 15 days covered the dish, formed small white to peach (No. 46) colored primordia, with small light yellow (No. 8-G) drops on the surface. After 15 days on MEA and 21 days on SDA mycelium has covered the dish, changing the color of the medium to reddish yellow (No. 12) on MEA, and mustard yellow (No. 9-H) to brick red (No. 19) on SDA. After 12 days incubation the mean surface covered by mycelium was 53.86 cm^2 on PDA, 51.92 cm^2 on MEA and 39.32 cm^2 on SDA (Tables 1 and 2).

Temperature 25°C. On PDA mycelium presented granular surface, covering the dish after 15 days and forming small white primordia which became peach (No. 46) colored, with light yellow (No. 8-G) drops on the surface. After 15 days mycelium has covered the dish and changed it to reddish yellow (No. 12) on MEA and to mustard yellow (No. 9-H) and brick red (No. 19) on SDA. In the latter medium after 15 days, light (No. 8-G) to mustard yellow (No. 9-H) aggregations as clusters began to form and dark brick red (No. 20) drops were observed at the

edge of the dish. On day 12th after inoculation the mean surface covered by mycelium was 56.89 cm^2 on PDA, 58.12 cm^2 on MEA and 53.59 cm^2 on SDA (Tables 1 and 2).

Temperature 27.5^{0}C. On PDA the mycelium density was greater at the center of the dish with a well marked concentric zone. On SDA well marked irregular concentric circles of dark brown (No. 18), pistachio green (No. 68) and wine (No. 76) were observed, in addition to dark brick red (No. 20) drops formed at the edges of the dish. When the entire dish had been covered, a reddish yellow (No. 12) was noted on MEA. On SDA the color of the medium changed to mustard yellow (No. 9-H) or brick red (No. 19). At day 12 of incubation the mean surface covered by the mycelium was 60.84 cm^2 on PDA, 54.63 cm^2 on MEA and 60.55 cm^2 on SDA (Tables 1 and 2).

Microscopic observations

On PDA, MEA and SDA the hyphae were from (1.2-)1.6-4(-4.8) µm in diam., thin walled, with dichotomous and acremoniform branching, with H shaped anastomosis, hyaline on PDA and MEA and yellowish red (No. 14) or reddish yellow (No. 12) on SDA. There were varicose hyphae on MEA and SDA. Terminal chlamidospores (9.6-) 10.4 x 12.8 µm, ovoid, thickwalled and reddish yellow (No. 12) on SDA were observed.

DISCUSSION

Maximum mycelial growth was observed at 27.5^{0}C on PDA and SDA and at 25^{0}C on MEA (Table 1). No significant differences were registered in regards to mycelium growth at 22.5 and 25^{0}C on PDA, 22.5 and 27.5^{0}C on MEA and 20 and 22.5^{0}C on SDA. On MEA the optimal growth range was wider, from 22.5 to 27.5^{0}C (Table 1). However, no significant differences were noticed among these culture media at 27.5^{0}C. On PDA, primordia were formed as observed by Nobles (1965), and Obregón-Arceo and Echenique-Manrique (1974), and were similar to those described by Findlay (1951). The pigmentation of MEA and SDA media was noticeable; Cartwright (1938) described it on malt agar. The sweet odor bserved in the three media, was also noted by Badcock (1939).

The chlamydospores formation was only observed on SDA, which agrees with Snell (1922), Nobles (1965) and Obregón-Arceo and Echenique-Manrique (1974). Septa and clamp connections were rare on PDA and SDA, but abundant on MEA. Cartwright (1938) mentioned that clamp connections are rarely found on culture media, although not rare in the mycelium in wood attacked by the fungus.

ACKNOWLEDGMENTS

The authors thanks to CONACyT for the support received (part of projects 0088N-N9106 and 1810-N9211). They also express their thanks to their colleagues Dulce Salmones, Manuel Juárez, Rosalia Pérez and Isabel Lara-Herrera for their help in the laboratory.

LITERATURE CITED

Badcock, E.C., 1939. Preliminary account of the odor of wood-destroying fungi in culture. *Trans. Brit. Myc. Soc. 23:* 188-198.

Cartwrigth, K.G., 1938. The relation between field and laboratory work in mycology. *Trans. Brit. Myc. Soc. 22:* 222-238.

Color Identification Chart, 1969. Royal Botanical Garden Edinburgh. *Flora of British Fungi*. Her Majesty's Stationary Office, London.

Dickinson, D.J., 1979. The decay of commercial timbers. *In*: J.C. Frankland, J.N. Hedger and M.S. Swift (eds.), *Decomposer basidiomycetes: their biology and ecology*. Cambridge University Press, Cambridge.

Dubovoy, C., T. Herrera, 1967. Estudio morfológico de los micelios de *Psilocybe caerulescens* Murrill. en diversos medios líquidos de cultivo. *Anales Inst. Biol. Univ. Nac. Autón. Mexico. Ser. Bot. 39:* 45-79.

Findlay, W.P.K., 1951. A note on the fungi of less common occurrence in houses. *Trans. Brit. Myc. Soc. 34:* 35-37.

Gómez-Nava, M.S., R. Echenique-Manrique, R. Salinas-Quinard, 1969. Índices de laboratorio sobre resistencia de la madera a la pudrición de once especies forestales mexicanas. *Bol. Téc. Inst. Nac. Invest. For. 31.*

Guzmán, G., 1977. *Identificación de los hongos comestibles, venenosos, alucinantes y destructores de la madera.* Limusa, México, D.F.

Nobles, M., 1965. Identification of cultures of wood-inhabiting Hymenomycetes. *Can. J. Botany 43:* 1097-1139.

Obregón-Arceo, M.C., R. Echenique-Manrique, 1974. Identificación de los hongos habitantes en postes de madera. *Anales Inst. Biol. Univ. Nac. Autón. México 45:* 11-20.

Snell, W., 1922. Studies of certain fungi of economic importance in the decay of building timbers. *Bull. U.S. Dept. Agric. 1053:* 1-47.

Stalpers, J.A., 1978. Identification of wood-inhabiting Aphyllophorales in pure culture. *Stud. Mycol. 16:* 1-248.

Suominen, J., 1973. On the occurrence of fungus *Lentinus lepideus* Fr. on railway sleepers in Finland. *Karstenia 13:* 40-43.

Table 1. Mean area of mycelium (cm^2) of *Lentinus lepideus* at 12 days of incubation.

TEMPERATURE (°C)	CULTURE MEDIA PDA $\bar{x} \pm \sigma$	MEA $\bar{x} \pm \sigma$	SDA $\bar{x} \pm \sigma$
20	45.28 3.13 a	44.53 0.79 a	34.23 4.48 a
22.5	53.86 2.54 b	51.92 1.54 b	39.32 3.67 a
25	56.89 2.23 b	58.12 3.41 c	53.59 1.24 b
27.5	60.84 2.59 c	54.63 8.16 bc	60.55 1.81 c

PDA: potato dextrose agar; MEA: malt extract agar; SDA: Sabouraud dextrose agar.
Different small letters after data indicate significant differences ($p<0.05$) according to Bonferroni's multiple comparison method.

Table 2. Behavior of *Lentinus lepideus* in the cultivation media at different temperatures.

Culture Media	Temperature	Mycelial Characteristics Color	Density	Texture	1	2	3
PDA	20	Wh	r	w	15	-	+
	22.5	Wh	r	w	15	-	+
	25	Wh	r	w	15	-	+
	27.5	Wh-SP	r	w	15	-	+
MEA	20	Wh-G	s	f	15	+	-
	22.5	Wh-G	s	f	15	+	-
	25	Wh-G	s	f	15	+	-
	27.5	Wh-G	s	f	15	+	-
SDA	20	Wh-S	r	f-w	18	+	-
	22.5	Wh-S	s	f	21	+	-
	25	Wh-S	s	f	15	+	-
	27.5	Wh-S	r	f	15	+	+

1: Days to complete coverage of Petri dish; 2: change of color in medium; 3: primordia.
Wh: white; SP: salmon pink; G: grey; S: sienna.
r: regular; w: wooly; s: scant; f: filmy.
+: present; -: absent.