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# CLASSICAL CHARACTERIZATION OF MUSHROOM GENETIC RESOURCES FROM TEMPERATE AND TROPICAL REGIONS OF MEXICO

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## ABSTRACT

**Native strains from temperate, tropical and subtropical regions of Mexico were studied in the laboratory. Strains belonging to the genera *Agaricus*, *Auricularia, Ganoderma*, *Lentinula*, *Neolentinus*, *Pleurotus* and *Volvariella* were characterized on potato-dextrose-agar medium (PDA) using petri dishes. Comparative characterization involved mycelial morphology, growth rate, residual reducing sugar and fresh biomass production. A strain of *Agaricus robustissimus* showed mycelial growth of high density, high growth rate of 0.188 mm/h, regular residual reducing sugar (56.0%), and high biomass production (4.7 g/L/day) in comparison with other strains of the same genus. A strain of *P. djamor* (CP-143) had high growth rate (0.185 mm/h), regular residual reducing sugar (66.0%), and high biomass production (3.7 g/L/day). Differing data were also recorded in *Auricularia fuscosuccinea*, *Ganoderma curtisii*, *G. lucidum*, *Lentinula boryana*, *Neolentinus lepideus*, and *Volvariella* spp. for growth rate (0.055-0.267 mm/h), residual reducing sugar (0-84.0%), and biomass production (1.45-3.72 g/L/day).**

**Key words: Edible mushrooms, genetic resources, germplasm characterization, fungal biomass, Mexico.**

## INTRODUCTION

The collection, characterization and conservation of mushroom genetic resources have become issues of scientific, biological and industrial importance worldwide, as well as a fundamental strategy for developing new generations of commercial strains[6]. Wild populations represent not only mushrooms to be potentially cultivated, but also a prominent source of biosynthetic products and genes for genetic engineering. Most global and regional efforts so far have been focused on the commercially important mushrooms *Agaricus* and *Lentinula*[1,2,5,15].

Latin America is a fundamental region for the conservation of the world's biodiversity. Six countries (Brazil, Colombia, Ecuador, Mexico, Peru, Venezuela) from this region are considered as real megadiversity places[4]. It is paradoxical, however, that there are only a few research programs for the recovery, characterization and conservation of native mushroom germplasm, wherein there has been a remarkable development of the mushroom industry in recent years[7].

We established a Centre of Mushroom Genetic Resources (CREGENHC) at COLPOS since 2004[8,14], and started studies on germplasm characterization [9,10,13,14]. In this study, selected Mexican germplasm maintained at CREGENHC was characterized describing mycelial morphology, growth rate, residual reducing sugar and fresh biomass production.

## MATERIALS AND METHODS

*Native germplasm*. Several species and strains of the genera *Agaricus* (7), *Auricularia* (1), *Ganoderma* (2), *Lentinula* (1), *Neolentinus* (1), *Pleurotus* (9), and *Volvariella* (1) were selected (**Table 1**). All strains are deposited at the Centre of Mushroom Genetic Resources (CREGENHC) at COLPOS, and were maintained and subcultured in potato-dextrose-agar (PDA, Sigma, U.S.A.). All species and their authorities are in accord with those from an international data base (www.indexfungorum.org).

*Reference strains*. Standard commercial strains were included in main genera for comparative analysis. This was so for *Agaricus* (*A. bisporus* var. *bisporus*, CP-39; *A. bitorquis*, CP-43 and CP-57 from Thailand); *Pleurotus* (*P. ostreatus*, CP-37 and CP-50; *P. ostreatus* f. sp. florida, CP-11; *P. pulmonarius*, CP-16 and CP-32); and *Lentinula* (*L. edodes*, CP-7).

*Colony morphology*. Main characteristics of mycelial colonies, such as density, texture, color, aerial hyphae, colony growth, and presence of exudates, were simple observations recorded after complete colonization of petri dishes.

*Fungal biomass and apical growth*. Biomass was determined as previously described[3] using a cellophane film on the agar surface of the culture medium under sterile conditions. An agar piece of inoculum (0.5 $cm^2$) from each strain was then placed onto the film at the center of the petri dish (9 cm diameter). After inoculation, petri dishes were incubated at 25 C for complete colonization. Fresh weight biomass was assessed by lifting away the mycelium grown on the cellophane film using sterile forceps. The colonized film was immediately weighed in an electronic analytical balance. The mycelial growth rate was measured considering colony diameter (mm) as a function of growth time (h) in petri dishes. All experiments were carried out with three replicates.

*Determination of pH and reducing sugar*. Initial pH was assessed at inoculation taking a sample of a complete agar plate.

In Erlenmeyer flasks, the agar plate was homogenized with 90 ml distilled water. Final pH was determined after lifting away the mycelium growing onto the cellophane film, taking a remaining agar plate to be homogenized with 90 ml distilled water. All readings were taken using a calibrated electronic pH meter (Knick, Germany). The concentration of residual reducing sugar was assessed according to the Miller method[11].

*Statistical analysis*. Data were subjected to standard ANOVA analysis and mean multiple comparisons using the Duncan test.

**Table 1**. Strains studied in this research, which are deposited at the Centre of Mushroom Genetic Resources (CREGENHC), COLPOS, Mexico.

| Species | Origin of strains (Country or State) | Number | Code |
|---|---|---|---|
| Native strains | | | |
| *Agaricus abruptibulbus* Peck | Puebla | 2 | CP-87, CP-140 |
| *A. augustus* Fr. | Mexico | 1 | CP-80 |
| *A. bitorquis* (Quél.)Sacc. | Puebla | 2 | CP-84, CP-85 |
| *A. osecanus* Pilát | Puebla | 1 | CP-83 |
| *A. robustissimus* Panizzi | Puebla | 1 | CP-73 |
| *Auricularia fuscosuccinea* (Mont.)Henn. | Chiapas | 1 | CP-103 |
| *Ganoderma curtisii* (Berk.)Murrill | Morelos | 1 | CP-145 |
| *G. lucidum* (Curtis)P. Karst. | Morelos | 1 | CP-158 |
| *Lentinula boryana* (Berk. & Mont.)Pegler | Veracruz | 1 | CP-5 (ATCC-90026) |
| *Neolentinus lepideus* (Fr.)Redhead & Ginns | Veracruz | 1 | CP-6 (ATCC-62610) |
| *Pleurotus cystidiosus* O.K. Mill. | Veracruz | 1 | CP-18 (ATCC-64764) |
| *P. djamor* (Rumph. ex Fr.)Boedijn | Mexico | 1 | CP-143 |
| | Michoacan | 1 | CP-53 |
| | Morelos | 3 | CP-34, CP-44, CP-76 |
| *P. levis* (Berk. & M.A. Curtis)Singer | Puebla | 1 | CP-30 |
| *Pleurotus* spp. | Chiapas, Veracruz | 2 | CP-91, CP-15 |
| *Volvariella* spp. | Puebla | 1 | CP-19 |
| Reference strains | | | |
| *A. bisporus* var. *bisporus* (J.E. Lange)Pilát | Spain | 1 | CP-39 |
| *A. bitorquis* (Quél.)Sacc. | Thailand | 2 | CP-43, CP-57 |
| *L. edodes* (Berk.)Pegler | Hong Kong | 1 | CP-7 |
| *P. ostreatus* (Jacq.)P. Kumm. | Germany, Mexico | 2 | CP-37 (ATCC-60271), CP-50 |
| *P. ostreatus* (Jacq.)P. Kumm. f. sp. florida | Germany | 1 | CP-11 |
| *P. pulmonarius* (Fr.)Quél. | Hong Kong, U.S.A. | 2 | CP-16, CP-32 |

ATCC= American Type Culture Collection, Manassas, Virginia, U.S.A.

## RESULTS AND DISCUSSION

*Agaricus*. A wide range of variation in colony morphology, growth rate, biomass production, and residual reducing sugar was observed in strains studied. Main characteristics regarding colony morphology are shown in **Table 2**. Mycelial colonies of regular and irregular growth were white and off-white. They showed high, regular and low density; velvety, floccose and hairy texture; and abundant, regular and scarce aerial hyphae. The strain CP-73 of *A. robustissimus* colonized the petri dish in 10 days (growth rate: 0.188 mm/h), while it took 20-42 days for the rest of native strains and 24-32 days for reference strains (**Table 3**). Previous work by the authors[9] reported a relatively high biological efficiency (45.9%) in this strain cultivated on compost. The slowest growth rates were recorded in strains CP-80 (0.020 mm/h) of *A. augustus* and CP-140 (0.024 mm/h) of *A. abruptibulbus*. The highest biomass production was 4.7 g/L/day for strain CP-73 (*A. robustissimus*), while the lowest was 0.656 g/L/day for strain CP-140. Biomass production in reference strains ranged from 0.781-1.09 g/L/day. Only the strains CP-83 of *A. osecanus* (27.2%), CP-85 of *A. bitorquis* (33.4%), CP-80 of *A. augustus* (34.0%), and CP-87 (40.1%) and CP-140 (45.8%) of *A. abruptibulbus* had less than 50% of the residual reducing sugar in the culture medium (**Table 3**). In the reference strains, only *A. bisporus* showed a residual reducing sugar lower than 50% (CP-39, 49.3%).

Other strains, CP-73 (56.0%) of *A. robustissimus*, and CP-43 (61.0%), CP-57 (57.8%) and CP-84 (80.0%) of *A. bitorquis* showed more than 55% of the residual reducing sugar. After mycelial growth, the final pH of the culture medium in reference strains (6.1-6.5) was higher than those from native strains which ranged from 5.7 (CP-80) to 5.9 (CP-83, CP-84, CP-140).

*Pleurotus*. Mycelial colonies of regular and irregular growth were white and pinkish white (**Table 4**). They showed high and regular density; cottony, velvety, and hairy texture; and abundant and regular aerial hyphae. Native strains colonized the petri dish in 7-42 days, showing a growth rate ranging from 0.028 mm/h in *P. levis* to 0.226 mm/h in the strain CP-15 of *Pleurotus* spp. (**Table 5**). Reference strains colonized the petri dish in 7-16 days, and their mycelial growth rate varied from 0.100-0.262 mm/h. The highest production of biomass of 3.95 g/L/day was recorded in the strain CP-32 of *P. pulmonarius*. The reference strain CP-11 of *P. ostreatus* f. sp. florida showed the lowest residual reducing sugar (3.7%) present in the culture medium. The lowest production of biomass (1.13 g/L/day) and the highest residual reducing sugar (80.0%) was recorded in the strain CP-18 of *P. cystidiosus*. In the strains of *P. djamor*, a common species in tropical and temperate regions, it was recorded a wide range of variation in growth rate (0.052-0.185 mm/h), biomass production (1.77-3.7 g/L/day), and residual reducing sugar (14.0-76.0%). After mycelial growth, the final pH of the culture medium ranged from 5.8 (CP-53) to 6.8 (CP-11) in all strains studied. The strain CP-30 of *P. levis* had white colonies of irregular growth, and was a slow growing strain in comparison with the reference strains. However, a residual reducing sugar of only 6.2% was recorded in this strain.

*Auricularia, Ganoderma, Lentinula, Neolentinus, Volvariella*. The colony morphology, residual reducing sugar, and biomass production of these mushrooms are shown in **Tables 6-7**. The strain CP-103 of *A. fuscosuccinea* had white dense colonies with pale brownish areas, velvety texture,

**Table 2.** Colony morphology of *Agaricus* strains grown on PDA medium at 25 C.

| Species | Code | Mycelial characteristics | | | | |
|---|---|---|---|---|---|---|
| | | Density | Texture | Color | Aerial hyphae | Colony growth |
| Reference strains | | | | | | |
| *A. bisporus* | CP-39 | +++ | Velvety | White | Scarce | Irregular |
| *A. bitorquis* | CP-43 | + | Velvety | Off-white | Abundant | Regular |
| *A. bitorquis* | CP-57 | + | Velvety | White | Regular | Regular |
| Native strains | | | | | | |
| *A. abruptibulbus* | CP-87 | +++ | Hairy | Off-white | Regular | Regular |
| *A. abruptibulbus* | CP-140 | + | Velvety | Off-white | Scarce | Irregular |
| *A. augustus* | CP-80 | +++ | Hairy | Off-white | Scarce | Irregular |
| *A. bitorquis* | CP-84 | ++ | Hairy | White | Regular | Irregular |
| *A. bitorquis* | CP-85 | ++ | Hairy | Off-white | Regular | Irregular |
| *A. osecanus* | CP-83 | ++ | Velvety | White | Abundant | Regular |
| *A. robustissimus* | CP-73 | +++ | Floccose | Off-white | Abundant | Regular |

+++= High density. ++= Regular density. += Low density.

**Table 3**. Growth rate and biomass production by *Agaricus* strains grown on PDA medium (initial pH= 5.8) at 25 C.

| Species | Code | Growth period (days) | Final pH | Growth rate (mm/h) | Residual reducing sugar (%) | Biomass fresh weight* (g/L/day) |
|---|---|---|---|---|---|---|
| Reference strains | | | | | | |
| *A. bisporus* | CP-39 | 32 | 6.3 | 0.026 | 49.3 | $0.781\pm0.00^{a,b}$ |
| *A. bitorquis* | CP-43 | 32 | 6.5 | 0.027 | 61.0 | $1.08\pm0.08^{c,d}$ |
| *A. bitorquis* | CP-57 | 24 | 6.1 | 0.036 | 57.8 | $1.09\pm0.10^{d}$ |
| Native strains | | | | | | |
| *A. abruptibulbus* | CP-87 | 20 | 5.8 | 0.049 | 40.1 | $2.13\pm0.23^{f}$ |
| *A. abruptibulbus* | CP-140 | 38 | 5.9 | 0.024 | 45.8 | $0.656\pm0.06^{a}$ |
| *A. augustus* | CP-80 | 38 | 5.7 | 0.020 | 34.0 | $0.702\pm0.02^{a}$ |
| *A. bitorquis* | CP-84 | 20 | 5.9 | 0.033 | 80.0 | $1.45\pm0.13^{e}$ |
| *A. bitorquis* | CP-85 | 42 | 5.8 | 0.044 | 33.4 | $0.88\pm0.03^{b,c}$ |
| *A. osecanus* | CP-83 | 24 | 5.9 | 0.062 | 27.2 | $1.4\pm0.06^{e}$ |
| *A. robustissimus* | CP-73 | 10 | 5.8 | 0.188 | 56.0 | $4.7\pm0.04^{g}$ |

*= Different letters in the same column indicated significant difference among values at level $\alpha= 0.05$, according to Duncan test.

**Table 4**. Colony morphology of *Pleurotus* strains grown on PDA medium at 25 C.

| Species | Code | Mycelial characteristics | | | | |
|---|---|---|---|---|---|---|
| | | Density | Texture | Color | Aerial hyphae | Colony growth |
| Reference strains | | | | | | |
| *P. ostreatus* f.sp. florida | CP-11 | +++ | Cottony | White | Abundant | Regular |
| *P. ostreatus* | CP-37 | +++ | Cottony | White | Abundant | Regular |
| *P. ostreatus* | CP-50 | +++ | Cottony | White | Abundant | Regular |
| *P. pulmonarius* | CP-16 | +++ | Cottony | White | Abundant | Irregular |
| *P. pulmonarius* | CP-32 | +++ | Cottony | White | Regular | Regular |
| Native strains | | | | | | |
| *P. cystidiosus* | CP-18 | +++ | Cottony | White | Abundant | Regular |
| *P. djamor* | CP-34 | ++ | Cottony | Pinkish white | Abundant | Regular |
| *P. djamor* | CP-44 | ++ | Velvety | Pinkish white | Regular | Irregular |
| *P. djamor* | CP-53 | +++ | Velvety | White | Regular | Irregular |
| *P. djamor* | CP-76 | ++ | Velvety | White | Regular | Irregular |
| *P. djamor* | CP-143 | ++ | Cottony | Pinkish white | Abundant | Irregular |
| *P. levis* | CP-30 | ++ | Hairy | White | Regular | Irregular |
| *P.* spp. | CP-15 | +++ | Velvety | White | Regular | Irregular |
| *P.* spp. | CP-91 | ++ | Hairy | White | Regular | Irregular |

+++= High density. ++= Regular density.

**Table 5**. Growth rate and biomass production by *Pleurotus* strains grown on PDA medium (initial pH= 5.8) at 25 C.

| Species | Code | Growth period (days) | Final pH | Growth rate (mm/h) | Residual reducing sugar (%) | Biomass* fresh weight (g/L/day) |
|---|---|---|---|---|---|---|
| Reference strains | | | | | | |
| *P. ostreatus* f.sp. *florida* | CP-11 | 11 | 6.8 | 0.100 | 3.7 | $2.09\pm0.57^{c,d}$ |
| *P. ostreatus* | CP-37 | 7 | 6.1 | 0.262 | 59.6 | $2.8\pm0.03^{e}$ |
| *P. ostreatus* | CP-50 | 11 | 6.5 | 0.158 | 40.5 | $2.68\pm0.01^{e}$ |
| *P. pulmonarius* | CP-16 | 16 | 6.0 | 0.109 | 5.0 | $1.31\pm0.03^{b}$ |
| *P. pulmonarius* | CP-32 | 9 | 6.3 | 0.145 | 38.6 | $3.95\pm0.22^{g}$ |
| Native strains | | | | | | |
| *P. cystidiosus* | CP-18 | 25 | 6.0 | 0.044 | 80.0 | $1.13\pm0.05^{b}$ |
| *P. djamor* | CP-34 | 20 | 6.0 | 0.055 | 14.0 | $2.18\pm0.20^{d}$ |
| *P. djamor* | CP-44 | 19 | 6.4 | 0.052 | 72.0 | $2.0\pm0.02^{c,d}$ |
| *P. djamor* | CP-53 | 19 | 5.8 | 0.092 | 76.0 | $2.87\pm0.21^{e}$ |
| *P. djamor* | CP-76 | 25 | 5.9 | 0.058 | 32.7 | $1.77\pm0.14^{c}$ |
| *P. djamor* | CP-143 | 9 | 6.3 | 0.185 | 66.0 | $3.7\pm0.56^{f,g}$ |
| *P. levis* | CP-30 | 42 | 6.3 | 0.028 | 6.2 | $0.594\pm0.07^{a}$ |
| *P.* spp. | CP-15 | 7 | 6.0 | 0.226 | 70.2 | $3.43\pm0.32^{f}$ |
| *P.* spp. | CP-91 | 24 | 6.0 | 0.054 | 11.8 | $1.9\pm0.14^{c,d}$ |

*= Different letters in the same column indicated significant difference among values at level $\alpha$= 0.05, according to Duncan test.

and abundant aerial hyphae. Its growth rate was 0.111 mm/h after 16 days, the biomass produced was 2.43 g/L/day, the residual reducing sugar was 22.8%, and the final pH of the culture medium was 6.3.

The colony morphology of *Ganoderma* strains was similar, except for texture (velvety, hairy) and mycelial growth (regular, irregular). The strain CP-158 of *G. lucidum* showed higher values of growth rate (0.065 mm/h) after 15 days, as well as biomass production (3.5 g/L/day), but a lower value of residual reducing sugar (6.7%) than the strain CP-145 of *G. curtisii*. The final pH of the culture medium in both species varied from 4.3-4.7.

The strain CP-5 of *Lentinula boryana* showed dense white colonies with brownish areas and exudates, as well as irregular growth. This species showed slower growth rate (0.055 mm/h) and higher residual reducing sugar (48.3%) after 19 days than the reference strain CP-7 of *L. edodes*. However, its biomass production (2.48 g/L/day) was higher. The final pH of the culture medium was acid for both species (3.6-4.2).

The strain CP-6 of *Neolentinus lepideus* showed velvety off-white colonies of scarce aerial hyphae, irregular growth, and a growth rate of 0.099 mm/h after 19 days. However, it showed high production of biomass (3.72 g/L/day) and no residual reducing sugar in the culture medium. The final pH of the culture medium was 3.8.

Off-white mycelial colonies of the strain CP-19 of *Vovariella* spp. showed brownish areas, which are associated to the production of chlamydospores. These colonies are characterized by high growth rate (0.267 mm/h) after 6 days, low biomass production (1.45 g/L/day), and high residual reducing

**Table 6**. Colony morphology of strains from *Auricularia*, *Ganoderma, Lentinula*, *Neolentinus* and *Volvariella*, grown on PDA medium at 25 C.

| Species | Code | Mycelial characteristics | | | | | |
|---|---|---|---|---|---|---|---|
| | | Density | Texture | Color | Aerial hyphae | Colony growth | Exudates |
| Reference strains | | | | | | | |
| *Lentinula edodes* | CP-7 | +++ | Velvety | White | Regular | Irregular | - |
| Native strains | | | | | | | |
| *Auricularia fuscosuccinea* | CP-103 | +++ | Velvety | White, pale brownish areas | Abundant | Regular | - |
| *Ganoderma curtisii* | CP-145 | +++ | Velvety | Off-white | Scarce | Regular | - |
| *G. lucidum* | CP-158 | +++ | Hairy | Off-white | Scarce | Irregular | - |
| *Lentinula boryana* | CP-5 | +++ | Velvety | White, pale brownish areas | Regular | Irregular | Brownish |
| *Neolentinus lepideus* | CP-6 | ++ | Velvety | Off-white | Scarce | Irregular | - |
| *Volvariella* spp. | CP-19 | + | Hairy | Off-white, brownish areas | Abundant | Irregular | - |

+++= High density. ++= Regular density. += Low density.

**Table 7**. Growth rate and biomass production by strains from *Auricularia*, *Ganoderma, Lentinula*, *Neolentinus* and *Volvariella*, grown on PDA medium (initial pH= 5.8) at 25 C.

| Species | Code | Growth period (days) | Final pH | Growth rate (mm/h) | Residual reducing sugar (%) | Biomass fresh weight* (g/L/day) |
|---|---|---|---|---|---|---|
| Reference strains | | | | | | |
| *Lentinula edodes* | CP-7 | 19 | 3.6 | 0.099 | 0.0 | 1.98±0.15[a] |
| Native strains | | | | | | |
| *Auricularia fuscosuccinea* | CP-103 | 16 | 6.3 | 0.111 | 22.8 | 2.43±0.23[b] |
| *Ganoderma curtisii* | CP-145 | 16 | 4.7 | 0.060 | 21.6 | 2.46±0.06[b] |
| *G. lucidum* | CP-158 | 15 | 4.3 | 0.065 | 6.7 | 3.5±0.15[c] |
| *Lentinula boryana* | CP-5 | 19 | 4.2 | 0.055 | 48.3 | 2.48±0.16[b] |
| *Neolentinus lepideus* | CP-6 | 19 | 3.8 | 0.099 | 0.0 | 3.72±0.25[c] |
| *Volvariella* spp. | CP-19 | 6 | 5.8 | 0.267 | 84.0 | 1.45±0.70[a] |

*= Different letters in the same column indicated significant difference among values at level $\alpha$= 0.05, according to Duncan test.

**Table 8**. Comparison of growth rate, biomass production, residual reducing sugar, and final pH recorded within genera studied (initial pH= 5.8).

| Genera | Growth rate(mm/h) | Biomass fresh weight (g/L/day) | Residual reducing sugar (%) | Final pH |
|---|---|---|---|---|
| *Agaricus* | 0.020-0.188 | 0.656-4.7 | 27.2-80.0 | 5.7-6.5 |
| *Auricularia* | 0.111 | 2.43 | 22.8 | 6.3 |
| *Ganoderma* | 0.060-0.065 | 2.46-3.5 | 6.7-21.6 | 4.3-4.7 |
| *Lentinula* | 0.055-0.099 | 1.98-2.48 | 0.0-48.3 | 3.6-4.2 |
| *Neolentinus* | 0.099 | 3.72 | 0.0 | 3.8 |
| *Pleurotus* | 0.028-0.262 | 0.594-3.95 | 3.7-80.0 | 5.8-6.8 |
| *Volvariella* | 0.267 | 1.45 | 84.0 | 5.8 |

sugar (84.0%). The final pH of the culture medium was 5.8.

In general (**Table 8**), the mushroom genera *Ganoderma*, *Lentinula* and *Neolentinus* were capable of reaching low residual reducing sugar (0.0-6.7%), which was associated to a lower final pH of the culture medium (3.6-4.7). The other genera, *Agaricus*, *Auricularia*, *Pleurotus* and *Volvariella*, also had residual reducing sugar (3.7-84.0%), but the final pH was similar to initial pH or higher (5.7-6.8). Higher values of biomass production (3.95-4.7 g/L/day) were associated to those

genera (*Agaricus*, *Pleurotus*) in which high biological efficiencies have been reported[12]. In these cases, higher growth rates led to high biomass production. The genus *Volvariella* showed the highest growth rate (0.267 mm/h), however, the biomass produced was low (1.45 g/L/day).

## LITERATURE CITED


1. Callac, P. 1995. Breeding of edible fungi with emphasis on the variability among French genetic resources of *Agaricus bisporus*. *Can. J. Bot.* 73: S980-S986.
2. Chang, S. T., H. S. Kwan and Y. N. Kang. 1995. Collection, characterization, and utilization of germ plasm of *Lentinula edodes*. *Can. J. Bot.* 73: S955-S961.
3. De Araujo, A. A. and S. Roussos. 2002. A technique for mycelial development of ectomycorrhizal fungi on agar media. *Applied Biochemical and Biotechnology* 98: 311-318.
4. Galindo-Leal, C. 2001. Design and analysis of conservation projects in Latin America: an integrative approach to training. *Conservation Ecology* 5(2): 16.
5. Kerrigan, R. W. 1995. Global genetic resources for *Agaricus* breeding and cultivation. *Can. J. Bot.* 73: S973-S979.
6. Labarère, J. and U. G. Menini. 2000. *Characterization, Conservation, Evaluation and Utilization of Mushroom Genetic Resources for Food and Agriculture*. FAO, Bordeaux. 240 pp.
7. Martínez-Carrera, D. 2002. Current development of mushroom biotechnology in Latin America. *Micol. Apl. Int.* 14: 61-74.
8. Martínez-Carrera, D., M. Bonilla, M. Sobal, A. Aguilar, W. Martínez and A. Larqué-Saavedra. 1999. A culture collection of edible mushrooms and its significance for germplasm preservation, breeding, and the development of mushroom cultivation in Mexico. *Micol. Neotrop. Apl.* 12: 23-40.
9. Martínez-Carrera, D., M. Bonilla, W. Martínez, M. Sobal, A. Aguilar and E. Pellicer-González. 2001. Characterisation and cultivation of wild *Agaricus* species from Mexico. *Micol. Apl. Int.* 13: 9-24.
10. Martínez-Carrera, D., P. Morales and M. Sobal. 1991. Sexual fruiting bodies from synnemata in *Pleurotus smithii*. *Micol. Neotrop. Apl.* 4: 9-18.
11. Miller, G. L. 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal. Chem.* 31: 426-428.
12. Smith, J. F., T. R. Fermor and F. Zadrazil. 1988. Pretreatment of lignocellulosics for edible fungi. Pp. 3-13. *In*: *Treatment of Lignocellulosics with White Rot Fungi*. Eds. F. Zadrazil and P. Reiniger. Elsevier Applied Science Publ., Barking.
13. Sobal, M., P. Morales, W. Martínez, D. N. Pegler and D. Martínez-Carrera. 1997. Cultivation of *Lentinus levis* in Mexico. *Micol. Neotrop. Apl.* 10: 63-71.
14. Sobal, M., P. Morales, M. Bonilla, G. Huerta and D. Martínez-Carrera. 2007. El Centro de Recursos Genéticos de Hongos Comestibles (CREGENHC) del Colegio de Postgraduados. *In*: *El Cultivo de* Pleurotus *en México*. ECOSUR-IE-UNAM-COLPOS, México, D.F. (in press).
15. Sonnenberg, A. S. M. 2000. Genetics and breeding of *Agaricus bisporus*. *Mushroom Science* 15: 25-39.