

A CHAMBER FOR FRUITING *VOLVARIELLA VOLVACEA* IN LABORATORY

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ABSTRACT: A chamber for fruiting *Volvariella volvacea* in laboratory. A chamber for fruiting thermophilic mushrooms in laboratory is described. This design allows an easy control of temperature, relative humidity (between 100% and 85%), CO₂ levels, air exchange and illumination. Moreover, a diagram with further modifications is described. These improvements enable greater control of temperature, relative humidity and air exchanges. They are focused on the use of a Peltier cell and allows mesophilic mushrooms growth.

Key Words: chamber, fruiting, Peltier, temperature, humidity, CO₂, mushroom, cultivation.

RESUMEN: Una cámara para la fructificación de *Volvariella volvacea* en el laboratorio. Se presenta el diseño de una cámara dirigida al cultivo de hongos termófilos en el laboratorio. Los sistemas de control permiten regular de manera sencilla la temperatura, la humedad relativa, entre el 100% y el 85%, y la concentración de CO₂ del aire, así como los intercambios de aire fresco y la iluminación de la cámara de cultivo. Respecto al cultivo de hongos mesófilos, incluimos un esquema de las modificaciones que permiten un mayor control de la temperatura, la humedad relativa y la ventilación. Las modificaciones se centran en el uso de una unidad de refrigeración, que en este caso es una célula de Peltier.

Palabras clave: cámara, fructificación, Peltier, temperatura, humedad, CO₂, setas, cultivo.

RESUM: Una cambra per a la fructificació de *Volvariella volvacea* en el laboratori. Es presenta el disseny d'una cambra dirigida al cultiu de bolets termòfils en el laboratori. Els sistemes de control permeten regular de manera senzilla la temperatura, la humitat relativa, entre el 100% i el 85%, i la concentració de CO₂ de l'aire, així com els intercanvis d'aire fresc i la il·luminació de la cambra de cultiu. Pel que fa al cultiu de bolets mesòfils, incloem un esquema de les modificacions que permeten un millor control de la temperatura, la humitat relativa i la ventilació. Les modificacions es centren en l'ús d'una unitat de refrigeració, que en aquest cas és una cèl·lula de Peltier.

Paraules clau: cambra, fructificació, Peltier, temperatura, humitat, CO₂, bolets, cultiu.

INTRODUCTION

Chambers for fruiting mushrooms are a useful tool for taxonomic and development studies, since they allow researchers to grow and fruit saprotrophic fungi under controlled climatic conditions. These chambers usually are costly, especially for laboratories located in developing countries. Moreover, in such countries, a growing need for isolation and selection of strains, exists. Recently, several designs of fruiting chambers have been reported. They allow some control of climatic parameters. Some papers concerning *Volvariella volvacea* (LI, 1981; THIELKE, 1981; LI, 1984; SALMONES & GUZMÁN, 1994) describe simple chambers in order to grow this mushroom. All these designs are functional. However, their ability to control climatic parameters, especially relative humidity and CO₂ levels, is low. Recently, our group has focused its research towards thermophilic and mesophilic edible mushroom strains characterisation. To afford such studies in special areas, we have designed two simple fruiting chamber models. These fruiting chambers look useful to improve mushroom culture technologies, since it makes possible to study what parameters are the optimum ones for growing edible and medicinal mushrooms.

We present the chamber model to grow thermophilic mushrooms we have used to study *Volvariella volvacea*. We also include a scheme of a fruiting chamber for mesophilic mushrooms. Finally, we add

some modifications that are needed to control air exchanges, useful to grow some mushrooms that like a low airflow (e.g. *Pleurotus* sp.).

MATERIALS AND METHODS

The fruiting chamber (Fig. 1) consists on a glass box, an aquarium, of 100 × 30 × 40 cm. The structure is coated with a 2 mm expanded high-density foam sheet, and the frontal side's insulation is removable in order to look inside. The cover has a hydrostatic joint. The climatic unit consists of a 40W insulated heater (Fig. 1, R1), two 12V DC computer fans (Fig. 1, F1-F2) and a humidifier. They are disposed inside a PVC tube, (90 mm Ø) hanging from the cover. The "U" shape of the distal part of this tube, which can be filled with water from outside through a silicon pipe, has a removable paper pad attached to the internal surface. Paper pad and water acts as a humidifier unit. A sensor that is wired to a thermostat (Fig. 1, T1) controls the heater. When needed, fresh air is introduced by a peristaltic air pump (Fig. 1, P). Incoming air is filtered through a sterilised 0,2 µm PTFE filter. The chamber is provided with an air exhaust (Fig. 1, AE). Substrate temperature can be increased by a 4m plastic-coated heating wire, 40W, that is extended into the substrate. It is controlled trough a thermostat (Fig. 1, T2). Temperature and humidity are monitored by a couple of thermometers, that act as a psychrometer.

RESULTS

Mass or container (autoclavable polypropylene bags or HDPE rigid boxes) cultivation can be afforded in this fruiting chamber. Substrate heater allows a quick temperature increase and to maintain substrate at a different temperature than chamber air's one. When spawn running is started inside the chamber, fan F1 and heater R1 are switched on and air pump P is switched off. In less than 20 minutes, a 100% HR is reached. To start primordia formation, F1 and R1 are stopped, and fan F2 and pump P are switched on. That reduces HR to approximately 85%, meanwhile air temperature and CO₂ levels drop. Once cold shock has been accomplished, heater R1 is switched on again to reach the desired temperature. Varying air pump P, we will control CO₂ levels in air (measurable through AE). In our studies, a 0,5-2 L/min air pump has been found to be suitable. By experience, to avoid temperature peaks that can damage spawn run, it is necessary to place the sensor T2 as near as possible to the heater. R2 should have low power; we suggest no more than 40 W. To avoid disturbances in the temperature control, it is better that fans (F1-F2) have a low internal resistance. Despite our chamber allows climatic control, it is important to place the fungarium away from heat and light sources.

DISCUSSION

The climatic chamber presented here has proved to be quite useful to grow tropical mushrooms, especially for *Volvariella volvacea*. Temperature and HR conditions are achieved quickly after the fungarium is switched on. Moreover, this system allows temperature and HR control between the growth parameters of this mushroom. Mushrooms other than tropical may require higher humidity control and/or to be grown at a temperature lower than ambient. In such cases, we suggest to use a variant of the design presented above (Fig. 2). This design incorporates a cooler, which can be either a chiller or a Peltier cell. The second option is more suitable for low volume fungaria as the one presented here.

The use of a Peltier cell allows to reduce air temperature, and therefore, to increase its relative humidity. To avoid problems with condensed water, the piece of tube where Peltier has been located should be slightly tilt in order to bring it back to the humidifier. CO₂ present in the incubation room comes from metabolic activity of the mycelia. In case it was necessary to get higher CO₂ levels since the beginning, it can be incorporated either from a gas cylinder, either produced in situ (STRAATSMA et al, 1986). In this case, to achieve a 0,5% CO₂ concentration in our fungarium filled with 40 litres of substrate, it is necessary to introduce 17,8 mmols Na₂CO₃ and 5% (w/v) HCl. Culture in containers placed inside the fungarium has proved to be more useful to avoid

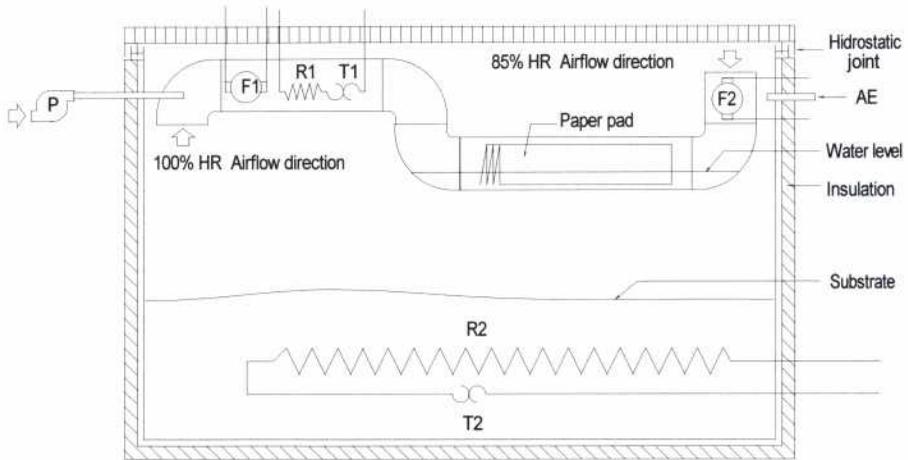


Fig. 1: Fruiting chamber for thermophilic mushrooms. P: Pump. F: Fan. T: Thermostat. R: Resistance (heater). AE: Air exhaust. Big arrows: Airflow direction.

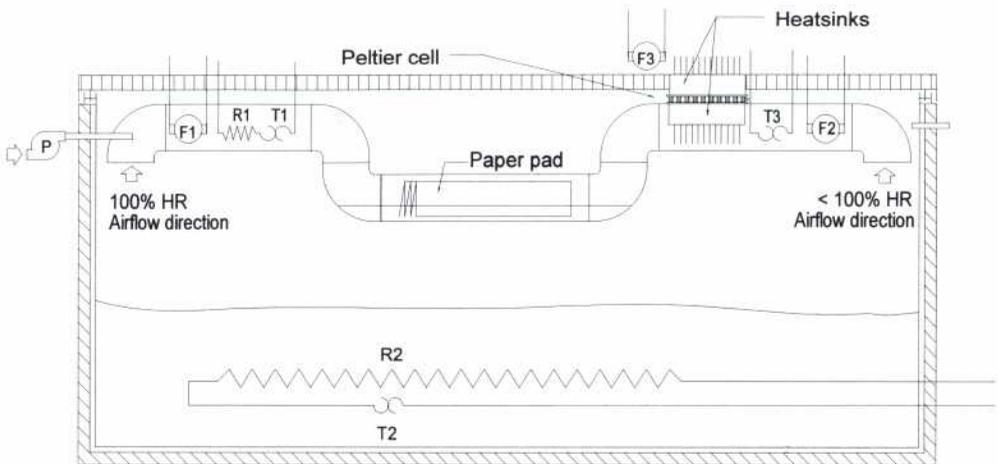


Fig 2: Fruiting chamber for mesophilic mushrooms. P: Pump. F: Fan. T: Thermostat. R: Resistance (heater). AE: Air exhaust. Big arrows: Airflow direction.

contamination, since handling is easier than in mass culture. In no case paper pad has been contaminated with fungi. Anyway, in order to prevent it became a source of problems; we have added H_2O_2 into the humidifier just before introducing the substrate.

Some mushrooms, like *Pleurotus ostreatus*, need a reduced airflow in the growing room. Therefore, a perforated 'ceiling' below the climatic unit should be added. A secondary duct also should be added, connecting the growing room and the climatic unit. Controlling returning airflow, we will manage incoming air in the growing room. Further modifications could allow substrate pasteurisation and inoculation in situ.

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BIBLIOGRAPHY

- LI, G.S. F. (1981). Simple techniques for fruiting of the straw mushroom *Volvariella volvacea* (Bull. ex Fr.) Sing. in laboratories. *Mush. Newsletter for the Tropics*, 2: 2-8.
- LI, G.S. F. (1984). A chamber for fruiting of *Volvariella volvacea* in laboratories. *Mush. Newsletter for the Tropics*, 4: 11-14.
- SALMONES, D. & GUZMÁN, G. (1994). Cámara para la obtención de fructificaciones del hongo comestible *Volvariella volvacea* en el laboratorio. *Revista Mexicana de Micología*, 10: 193-198.
- STRAATSMA, G., L.J.L.D. VAN GRIENSVEN & J. BRUINSMA. (1986). Root influence on in vitro growth of hyphae of the mycorrhizal mushroom *Cantharellus cibarius* replaced by carbon dioxide. *Physiol. Plant*, 67: 521-528.
- THIELKE, C. (1981). Die Fruktifikation der Kulturform von *Volvariella volvacea* (Bull. ex Fries) Sing. unter Laboratoriumsbedingungen. *Der Champignon (Bonn-Bad Godesberg)*, 238: 25-26.