

## The Exploitation and Cultivation of *Pleurotus nebrodensis* in China

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**Abstract:** As a new species of edible mushroom, *Pleurotus nebrodensis* has been paid more and more attention in recent years for its particular flavor, ample nutrition and medicinal value. In 2002, the gross yield in China was 34,325 tons. *P. nebrodensis* sometimes shows autoecious characteristics and broad adaptability to nutrition. The optimum temperature for mycelial growth is 25°C, 0-13°C for differentiation and 8-13°C for growth of fructification, respectively. The optimum water content of the substrate for mycelial growth is 60-70%, and the optimum relative humidity of air for fruit body differentiation and growth is 87-95%. Mycelial growth does not need light, while fruit body differentiation and growth needs dispersed light. Fresh air is needed both for the growth of mycelium and fruit body. The optimum pH for mycelial growth is 5.5-6.5. At present, there are three types of cultivating modes, viz. industrial production, out-of-season production using refrigeration or caves and seasonal establishment cultivation. Substitute cultivation using plastic bags (17cm in width, 34cm in length and 0.004cm in thickness) is widely adopted and artificial button reduction is one of the important techniques in mushroom production. The best time for harvesting *P. nebrodensis* is when the pileus has opened evenly and the brim is still involute. Commercial treatments of *P. nebrodensis* include pre-cooling, refitting, grading, packing and keeping fresh during storage and transportation. The commercial types of *P. nebrodensis* include fresh mushroom, kept-fresh mushroom, pickled mushroom, canned mushroom and dry slices. As a new type of exploited edible fungus, *P. nebrodensis* has great market potential and an expansive development future.

**Key words:** *Pleurotus nebrodensis*, biological characteristics, cultivating modes, commercial treatments of fresh mushroom

### 1 Introduction

*Pleurotus nebrodensis* is a new type of commercially cultivated edible mushroom. It is well favoured by consumers for its fat body, pure white color, crisp and tender character, delicious taste and ample nutrition. In 1983, when researching and domesticating wild *Pleurotus ferulae* in Tuoli, Xinjiang, Cao Yuqing and Mou Chuanjing observed that there were different cultural characters between the detached strains - K001, K002 and K005. In 1986, they collected a specimen of K111 in Mulei, Xinjiang and, on further study, found that there was a distinct difference in the outer form and cultural characters between *P. ferulae* and K005 and K111. Through identification, K005 and K111 were named *Pleurotus eryngii* (DC.ex.Fr.) Quel.var.tuoliensis Mou. n. var. After this, *P. ferulae* and *P. eryngii* (DC.ex.Fr.) Quel.var.tuoliensis Mou. n. var were popularized and cultivated. After the spawn was introduced from Mulei, Xinjiang and cultivated successfully on a large scale by Beijing Jinxin Edible Fungus Ltd in 1997, Bailing mushroom, the commercial name of white *P. eryngii*, was recommended and broadly accepted. Before and after this, it was named *P. nebrodensis* by Huang Nianlai and Mao Xiaolan. It was also pointed out by Huang Nianlai that the *P. eryngii* (DC.ex.Fr.) Quel.var.tuoliensis Mou. n. var was close to *P. nebrodensis*. They may be the same species with different names. In China, large-area cultivation of *P. nebrodensis* began from Beijing and developed in Xinjiang, Henan, Tianjin, Shandong, Hebei and Gansu. The statistical figures of Chinese Association of Mushroom showed that the gross yield of fresh *P. nebrodensis* in China was 34,325 tons in 2002.

### 2 Classification Status

*P. nebrodensis* belongs to *Pleurotus*, Pleurotaceae, Agaricales, Hymenomycetes, Basidiomycotina and its scientific name should be *P. eryngii* (DC.ex.Fr.) Quel. var. *nebrodensis* inzenga=*P. nebrodensis* or *Pleurotus nebrodensis* (inzengae) Quel. It also has numerous Chinese names including baiaweiwo, baiaweiceer, bailingceer, ciqinceer, baisebianzhong, bailinggu, baochigu, aweiceer and tuolibianzhong.

### 3 Distribution and Morphological Characters

Wild *P. nebrodensis* is distributed in Tuoli and Mulei, Xinjiang and northwest Sichuan, and is born out of the stalk bottoms or roots of umbelliferae herbage, such as asafetida, Xinjiang asafetida and eryngo. The pileus shows a spoon or umbrella shape with an area of 5-15cm × 9-15cm and white in color with cream colored speckles. Also, the brim of the pileus involutes, has a projecting middle part and is not sticky. The pileus grows evenly with a squama on the surface and chaps. The flesh is white and thick. The lamellae are sparse and 1-2mm apart. The stipe is a thin, long and solid column (7-11cm × 1-3cm). The spore is colorless, contains oil drip and has an elliptical appearance (8.75-13.75µm × 5-6.25µm).

The cultivated fruit body is fat and big with a sunken or even bottom and has a flat ball shape initially and then grows flat gradually. It has a scarce brim or no back brim, and has a pure white appearance. On its surface, it is close to satin or may be sometimes downy. A ripe pileus has a diameter of 5-15cm or more. The flesh is thick, white and does not change color. The lamellae are white and become a little yellow later. The adnation stipe is rarely excentric, wide at the upper end and thin at the bottom, is unshaped, crisp and solid with a length of 3-8cm and a width of 2-3cm. The basidium has four little peduncles and the spore is colorless and satin, contains oil drip and has an elliptical columnar appearance (9-13.5µm × 4.5-5.5µm). In late autumn, winter or early spring, fruit bodies appear on artificial substrates either singly or in clusters.

As a result of differences among strains and the environment during the course of cultivation, there are three types of commercial hymenium of *P. nebrodensis*, viz. the type with fan-shaped pileus and adnation stipe, the type with long adnation or excentric stipe and small clevis pileus, and the type with the bread pileus and near central stipe.<sup>[3]</sup> In addition, there are still diversified malformed mushrooms with anomalous shape.

### 4 Nutritional Components and Edible Value

*P. nebrodensis* is a type of fungus with ample nutrition and great edible value. Analytical data suggest that the ripe fruit body contains 89.24% water, 2.92% crude protein (27.14% in the dry mushroom), 1.725% amino acids (E/T 51.01%), 1.11% coarse-fiber, 0.68% ash, and the mineral elements include P, K, Na, Ca, Mg, Fe, Zn, Mn, Cu, Co, Ni, Se and Se. As for heavy metals, the contents of As, Hg, Pb and Cd are lower than 0.001, 0.00278, 0.39 and 0.03 mg/kg, respectively. They are all lower than the limits introduced in the Health Standard (GB7096-2003) and the industry standard of pollution-free established by Ministry of Agriculture (such as pollution-free food *Pleurotus sapidus* NY5096-2002). Among the proteins in the hymenium of *P. nebrodensis*, there are seventeen kinds of amino acids, and the content of eight essential amino acids are 0.88%-1.16%. The ratios of contents of essential amino acids to total amino acids in the mushroom budding period, growth period and ripening period are 44.1%, 49.05% and 51.01%, respectively. It is reported that the total content of essential amino acids among the proteins in fresh hymenium of *P. nebrodensis* is higher than that of *Hypsizigus marmoreus*, *Pleurotus* spp and the FAO/WHO standard at 4.08%, 7.96% and 24.00%, respectively. The amino acids and chemical grade of the proteins are higher than those of *H. marmoreus* and *Pleurotus* spp at 42.863% and 36.25%, and 37.36% and 26.52%, respectively. The exponent and biological value of essential amino acids are higher than that of *H. marmoreus* and *Pleurotus* spp at 10.24% and 9.96%, and 11.85% and 11.55%, respectively. However, the nutrition exponent of proteins is lower than those of *H. marmoreus* and *Pleurotus* spp. The ratio

coefficient of the amino acids is lower than that of *Pleurotus* spp. but higher than *H. marmoreus*.

## 5 Medicinal Components and Effects

As a medicinal mushroom, *P. nebrodensis* is recorded to cure numerous ailments including stomach cold and limb anaesthesia. Recent research has suggested that the content of crude polysaccharide in the dry hymenium of *P. nebrodensis* is as high as 6.015-7.04% and in mycelium 4.53%. It was found that the crude polysaccharide contained in hymenium and mycelium of *P. nebrodensis* has immunological activity. After extraction from the hymenium using hot water, PFW fraction, a type of polysaccharide, was deproteinized and chromatogrammed to produce PF1 and PF2 fractions. Through celiac injection into the mouse, the polysaccharides were found to inhibit S-180 tumor. Through a pilot study of antioxidation, different components extracted from the hymenium of *P. nebrodensis* were also found to remove oxygen radicals created in the Fenton reaction. Among the components, those extracted by acetic acid ethyl ester had the highest efficiency. Therefore, there may be new source of natural anti-oxidants in the hymenium of *P. nebrodensis*. PFA, another type of water-soluble polysaccharide extracted from the water-soaked hymenium of *P. nebrodensis* using 3% trichloroacetic acid, was deproteinized, dialyzed and precipitated to produce PF3 polysaccharide.

## 6 Biological Characteristics

### 6.1 Nutrition requirements

In nature, *P. nebrodensis* is mainly born out of large umbelliferae herbage, such as eryngo, and asafetida. It was once thought that *P. nebrodensis* is an autoecious fungus that could not be artificially cultivated. However, it was later shown that *P. nebrodensis* is saprophytic that occasionally shows some autoecious characteristics. Though the *P. nebrodensis* produced in Xinjiang is born out of the stalk bottoms or roots of asafetida, it was later shown that the mushroom can grow naturally, produce hymenia and complete the life cycle. Experiments have also indicated that *P. nebrodensis* has broad nutritional adaptability. Mixed in certain proportions, cotton-seed shuck, wood scraps, corn straw, peanut stalks, xylose dregs, sugarcane dregs, straw, cotton boll shuck and legumina can all be used as the main materials for the production of *P. nebrodensis*. Furthermore, bran, corn flour, rice chaff, urea, superphosphate, potassium dihydrogen phosphate, gypsum, calcium phosphate, yeast and saccharose can all be used as auxiliary materials to supply nitrogen and other nutrients.

### 6.2 Temperature

*P. nebrodensis* prefers middle or low temperatures and 25°C is optimal for mycelial growth. At 28°C and 26.5 °C, the mycelial growth rate decreases slightly, and decreases dramatically at 23.5°C. Mycelial growth ceases at 35°C. In the late period of mycelial growth, the temperature should be maintained at 0-13°C in order to promote reproductive growth. The hymenium grows rapidly at 15-18°C and the mushroom growing at 8-13°C has the best quality. However, the hymenium will turn yellow and decompose at 20°C.

### 6.3 Humidity

The mycelium will grow when the ratio of solids to water in the substrate is within the range of 1.1-2.6. Thus, the mycelium grows most rapidly when the water content of the substrate is in the range 60-70%. When the relative humidity of the air is between 87-95%, the hymenium grows normally, but at low temperatures (6-7°C) and in dry conditions, the surface of the pileus chaps frequently.

## 6.4 Light

During mycelium growth *P. nebrodensis* does not need light. However, when differentiating, the mushroom bud needs dispersed light. Also, the hymenium grows normally at a light intensity of 200-500 lux.

## 6.5 Aeration

When growing, the mycelium and hymenium of *P. nebrodensis* need fresh air so as to obtain sufficient oxygen. In airless growing rooms, malformations are often observed with some hymenia having a toadstool or crown shape.

## 6.6 pH value

In nature, *P. nebrodensis* grows on the roots of asafetida and the earth is alkaline with a pH value of 7.85. Research data indicate that the mycelium of *P. nebrodensis* can grow on substrates with pH values ranging from 5-11, but the optimum pH is 5.5-6.5.

## 7 Cultivation

### 7.1 Cultivation modes

Although large-scale commercial cultivation of *P. nebrodensis* commenced only about 10 years ago in China, there are presently three types of cultivation modes, viz. industrial production, anti-season production using refrigeration or caves, and seasonal establishment cultivation. The fresh mushrooms in the market are mostly offered through the way of seasonal establishment cultivation in late autumn, winter or early spring. Out-of-season production mostly offers fresh mushroom during the period from June to September, but the yield is limited. Although there are only relatively few manufacturers undertaking industrial production, they can frequently offer fresh mushrooms and further development in this direction is underway.

### 7.2 Spawn breeding

In terms of spawn breeding, there is no difference between mother spawn material and cultivation spawn. Potato dextrose agar is selected for mother spawn production, while cornmeal mash is used for producing wild and cultivation spawn. The mycelium of *P. nebrodensis* grows slower than *P. sapidus* and is easily contaminated. Therefore, it is important to apply strict controls. In practice, to obviate mycelium ageing, the spawn should be used immediately after the substrate has been fully colonized by the mycelium.

### 7.3 Technical essentials in seasonal establishment cultivation

Generally, spawn making and inoculation are carried out during the period from the last ten days of August to the end of September and then the mycelium is bred. After the mycelium has grown ripe, the new mushrooms are harvested during the period from October in the same year to May of the next year. If combined outdoor and indoor cultivation (mushroom sheds) was adopted, the production period will be shortened and new mushrooms will be harvested ahead of schedule. Preferably, mushrooms should be harvested in sunlit greenhouses and plastic sheds, although different shaped half-underground mushroom sheds are used in the drought-ridden and cold northwest areas of China.

The substrate is generally fermented through deposition, bagged and sterilized. Plastic bags of 17cm width,

34cm length and 0.004cm thickness are recommended and wet substrate of 1kg weight is bagged so that the size of single mushrooms accords with the market demands.

During commercial cultivation, artificial button induction is one of the important techniques. Using this technique, the time that the mushroom is ready for market can be controlled, and the mushroom will grow trimly and is thus easy to manage. At present, only one batch (flush) of mushrooms is harvested and the biological efficiency is as high as 30%. If the cultivated model of "spawn bury" or "spawn walling" is adopted, two batches of mushrooms are harvested and the biological efficiency is as high as 60%. It takes about 10-15 days from the appearance of buttons to harvesting although this time will be longer in the cold season.

## 8 Commercial Treatments, Storage and Transportation of Fresh Mushrooms

### 8.1 Harvesting

As a quality type edible fungus, fresh *P. nebrodensis* needs to be treated commercially so as to accord with the market. Therefore, harvesting is the key basic step to ensure quality and prolonged shelf life. The best time for harvesting is when the pileus has open evenly and fully and the brim is still involuted. When harvested, a first refitting is performed; i.e. the eye winkers adhering to the surface should be cleared away and the stipe cut evenly at the joint of the stipe and pileus. Then, the fresh mushrooms are placed into foam boxes according to different standards of size, shape and weight.

### 8.2 Pre-cooling

After harvesting, trimming and classification, the boxed fresh mushrooms should be placed under refrigeration at 0-1°C for 15-20h. When the central part of the mushroom body has been cooled to the same temperature as the refrigerator, i.e. fully cooled, the pre-cooling is completed.

### 8.3 Refitting and classification

There are strict requirements relating to the grading of *P. nebrodensis* in the market and the prices of the different grades of mushroom are very different. Generally, *P. nebrodensis* is classified into five grades. Grade A mushrooms are white or ivory color, fan-shaped, flat and have an unwrapped pileus with involuted brim, smooth surface, no gaps, no yellowing or waterlogged spots and no flaws. There is no villiform mycelium at the joint of the stipe and pileus, and the thickness of the central part of pileus is above 2.5cm. The stipe is cut away completely, and there are intact, clear veins and regular lamellas. The weight of a single mushroom ranges from 125g to 200g. Compared to Grade A, there is hollowness at the joint of the stipe and pileus of Grade B mushrooms, the pileus lacks smoothness and there are a few yellow spots or villiform mycelium. Compared with Grade A, there is no difference except that the weight of single mushroom of Grade C ranges from 50g to 124g. Similarly, there is no difference between grade D and grade B except that the weight of a single mushroom of grade D ranges from 50g to 124g. As for mushrooms of grade E, the pileus is twisted or malformed, the stipe is long with a small pileus, there are gaps or no involuted brim on the pileus, and no strict requirements with regard to the weight of single mushroom.

A second refitting should be carried out when grading so as to make the mushroom body more neat and beautiful.

### 8.4 Packing

After pre-cooling, refitted for a second time and graded, the kept-fresh mushrooms are packed. Most importantly, the mushrooms should be wrapped with wrappers having a 27cm × 27cm area. The wrappers should be in

accord with the demands of QB1014 food-wrapper and QB11680 food-wrapper health standards. The wrapped mushrooms should be placed into foam boxes one by one in layers with a 5kg net weight per box, and the box is then sealed with adhesive tape. The foam box should be in accordance with the health standards of food boxes and have dimensions of 48cm × 32cm × 20cm.

### 8.5 Keeping the mushrooms fresh during storage and transportation

The fresh *P. nebrodensis* are sold mostly in big cities and transported by air. They should be transported to the terminal markets within 24 h of being packed. The pre-cooled kept-fresh mushroom can be stored below 0°C and the temperature should be constant during the storage period. Generally, the kept-fresh mushroom can be stored for up to 3 months.

## 9 Market and Development Potential of *P. nebrodensis*

### 9.1 Commercial classification

There are five types of commercial *P. nebrodensis* including fresh mushroom, kept-fresh mushroom, pickled mushroom, canned mushroom and dry slices. Therefore, fresh *P. nebrodensis* is the form that is directly transported to the market for sale after only a little refitting. The kept-fresh form is the commercially treated fresh mushroom, which can be stored for long periods and be transported long-distances so that the value is increased. The pickled mushroom is stored in a saturated salt solution after certain technical treatments. The canned form is sealed in containers using standard can-making techniques, and can be stored for long periods. Dry slices are cut dried mushroom slices prepared according to specified standards, and can be most easily stored and transported.

### 9.2 Market demands for *P. nebrodensis*

The fresh mushroom, sold in all cities at a relatively low price, is suitable for low-earning employees and townspeople. The kept-fresh form, sold to wholesale markets in big cities or exported, is suitable for quality restaurants or hotels and demands a higher price. The pickled form, a mixture of good and poorer quality, is consumed by hotels and restaurants and is also the raw material for the canned form. The canned form, sold in big cities or exported, is a quality type edible fungus that has a higher price. However, there is little demand for dry slices, and more forms of marketing the mushroom will be developed in the future.

### 9.3 Development potential for *P. nebrodensis* production

Although prices fluctuate greatly, *P. nebrodensis* is still favored by culturists, proprietors and consumers and is praised as the "evergreen tree" for its characters: i.e. 1. a pleasing shape, fit for frying, cooking, stewing and as "maigre abalone"; 2. tasty, refreshing, buccal sense like abalone; 3. pure white in color; 4. storage qualities and ease of keeping fresh; 5. ease of storage and transportation. Therefore, as a new exploited type of edible fungus, *P. nebrodensis* has broad market appeal and developmental potential. Yields and consumption of this mushroom are likely to catch up with and exceed those of *Agaricus bisporus* and *Lentinula edodes*.