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Shelf life extension of white mushrooms (*Agaricus bisporus*) by low temperatures conditioning, modified atmosphere, and nanocomposite packaging material



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ABSTRACT

In this work, we have explored a new integrated approach for the shelf life extension of button mushrooms (*Agaricus bisporus*). The effect of temperature (4 °C and 25 °C), packaging configuration (PET/coating/LLDPE oxygen barrier material over conventional PVC stretchable film), and modified atmosphere (15% $O_2/5\%$ $CO_2/80\%$ N_2 over air) were monitored during 10 days of storage. The influence of a chitosan coating deposited on the cap surface was also investigated. Temperature was the most important factor in preserving the quality attributes of mushrooms over time. The test material had a positive impact on weight loss, cap opening percentage, and firmness of mushrooms compared with the control film ($\sim1.0\%$ versus $\sim7.1\%$; $\sim55\%$ versus $\sim65\%$; and ~10.3 N versus ~7.6 N, respectively), which was ascribed to the excellent and good oxygen and water vapor barrier properties of the new material, respectively. Mushrooms packaged under the modified atmosphere behaved decidedly better after a prolonged storage time of 22 days at 4 °C. Impressively, after this extended temporal window, the mushrooms looked freshly packed by fully recovering their original color. We explained this striking observation in consideration of the oxygen that permeated the package during these additional 12 days of storage, which would have promoted a gradual resumption of respiratory activity in the overall metabolism of the mushrooms after the "freezing" effect of the rich-CO₂ atmosphere inside the package.

1. Introduction

High nutritional value, sensory properties, medicinal attributes, ease of harvesting, and lower price compared to other mushrooms are the main reasons for the widespread cultivation of Agaricus bisporus (also interchangeably known as button mushrooms, white mushroom, and champignon) in many parts of the world, insomuch as it is currently the most cultivated edible mushroom worldwide (Meng et al., 2017; Qin et al., 2015). However, the commercial potential of this type of mushroom is somehow relented by its very short shelf life, which is ~3-4 days at room temperature and ~8 days under refrigerated conditions (Jiang, 2013). This short shelf life, especially if compared with other fresh vegetables, has a main structural reason: button mushrooms have no cuticle to act as a physical barrier against mechanical damage, water loss, or microbial attack. A high respiration rate and high moisture content contribute to the rapid senescence of button mushrooms, promoting microbial attack and enzymatic browning (Aguirre, Frias, Ryan, & Grogan, 2008). Eventually, color changes, tissue rotting, loss of turgor, off-flavors, and microbial spoilage become the most important quality attributes affecting postharvest storage, marketability at retail stores, and consumers' acceptance.

Different postharvest approaches have been proposed to control (and possibly delay) the rapid quality decay of button mushrooms. First, storage in refrigerated conditions relents overall metabolism, although it has been pointed out that this can also have detrimental effects on product quality, particularly during prolonged storage periods (Lagnika, Zhang, & Mothibe, 2013). Chemical pretreatment of button mushrooms using citric acid, ethylenediaminetetraacetic acid (EDTA), hydrogen peroxide, or sodium hypochlorite has been proposed by several authors, although undesirable changes in the appearance and general quality of the final product may occur (Lagnika et al., 2013).

Unconventional approaches have also been proposed to slow down the postharvest decay of button mushrooms, such as γ -irradiation (Benoit, D'Aprano, & Lacroix, 2000), ultrasound and high-pressure argon (Lagnika et al., 2013), pulsed light (Oliu, Aguayo, Belloso, & Fortuny, 2010), UV-c (Wu et al., 2016), gaseous ozone treatments (Akata, Torlak, & Erci, 2015), the use of edible coatings (Jiang, 2013), and antimicrobial and moisture-absorbing active

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packaging (Mahajan, Rodrigues, Motel, & Leonhard, 2008; Qin et al., 2015).

Packaging plays a crucial role in the control of the rate of mushrooms' senescence. Modified atmosphere packaging (MAP) in particular is a powerful tool to control both microbial growth and physiological effects in mushrooms (Li et al., 2014). In this regard, high CO2 concentrations have to be discouraged because anaerobic conditions can lead to metabolic disorders and undesirable fermentation resulting in off flavors (Jacxsens, Devlieghere, & Debevere, 2002). Nevertheless, high CO₂ concentrations (95-100%) in combination with ventilation using a new packaging method have been very recently proposed (Lin et al., 2017). Other authors have agreed on optimal recommended atmosphere with low O₂ content (less than 10%) and limited CO₂ content (5% maximum). However, combinations of O2 and CO2 are rather difficult to maintain over time because high gas permeability values and high perm-selectivity of packaging materials (i.e., a high ratio between CO2 and O2 permeability) would be needed (Guillaume, Schwab, Gastaldi, & Gontard, 2010). High O2 atmospheres have also been tested for button mushrooms. Liu, Wang, Zhu, and Wang (2010) reported that a high oxygen atmosphere, especially 100% O2, was a suitable method of storage for button mushrooms, whereas Liu and Wang (2012) demonstrated that mushrooms exposed to high oxygen concentration (80% O2) had a higher whiteness index and a lower increase in relative electrolyte leakage rate, lipid peroxidation, and ROS (O2. and H2O2) production indicating less membrane damage.

Both packaging technology and the selection of packaging material can have a dramatic impact on quality. Different materials can be selected in relation to storage conditions (refrigerated or room temperature), type of presentation (whole or sliced), and packaging technology (with or without MAP, type of MAP). Button mushrooms are conventionally packaged in rigid plastic (e.g., polyethylene terephthalate, PET) punnets or foam trays (e.g., expanded polystyrene, EPS) wrapped with PVC film or other stretchable films. However, alternatives have been proposed, such as the use of PET with different degrees of perforation (Taghizadeh, Gowen, Ward, & O'Donnell, 2010), biaxially oriented polypropylene (BOPP) (Xing, Wang, Feng, & Tan, 2008), and materials obtained from renewable resources such as poly(lactic acid) (PLA)/poly(ε-caprolactone) (PCL) blend films (Qin et al., 2015) and wheat gluten-coated paper (Guillaume et al., 2010).

In this work, we have investigated the combined effect of temperature, MAP, and packaging material on the shelf life extension of whole button mushrooms. In particular, we have selected an innovative bio-hybrid packaging material based on a "nano" technology with super oxygen barrier properties, but permeable to ${\rm CO_2}$ to a certain extent, in both dry and refrigerated conditions. We have also decided to test a relatively high O2 concentration (three times higher than CO2). The rationale underlying this approach is that such configuration would allow mushrooms to preserve their original quality attributes for a long time due to a twofold effect: at the beginning, the high oxygen concentration inside the package would act as a reservoir for the metabolism of the mushrooms; in a second step, as soon as the metabolism of the mushrooms decreases, the CO₂ accumulation inside the package would act as a preservative against the detrimental decay reactions that would otherwise impair mushrooms' marketability. Nonetheless, due to the increasing demand for replacing chlorine-based materials (such as PVC) with less impacting materials (PVC poses serious concerns due to the production of dioxin during incineration), the use of an alternative packaging configuration can also be seen in terms of environmental and consumers' health impact.

The effect of the deposition of a chitosan coating on the mushrooms' surface was also investigated. To the best of our knowledge, a similar approach has never been reported.



Fig. 1. Button mushrooms packaged in the bionanocomposite-based laminate/EPS tray (a) and in the conventional PVC stretchable/EPS tray configuration (b) tested in this study.

2. Materials and methods

2.1. Materials

Button mushrooms were kindly supplied by Fungorobica srl (Cenate Sotto, Italy). Mushrooms from second flush and at the closed cap stage were carefully selected according to a uniform shape and size (cap size of 40–50 mm diameter). Samples were then stored at 4 $^{\circ}$ C and 75 \pm 2% RH for 24 h before analyses.

A PET/coating/LLDPE film was used as a test packaging material. It was obtained by first depositing an oxygen barrier coating (0.5 μm thick) onto the 12 µm thick corona-treated PET film (Metalvuoto spa, Roncello, Italy), and then laminating the coated PET with the $60\,\mu m$ LLDPE layer (Metalvuoto spa, Roncello, Italy) by means of a doublecomponent polyurethane adhesive (AD 737, Novachem Industriale, Legnano, Italy). The oxygen barrier coating has been obtained according to the procedure reported in detail in our previous work (Introzzi et al., 2012). Briefly, it consists in a bionanocomposite coating made of a main biopolymer phase (the exopolysaccharide pullulan) that intercalates an inorganic filler (natural cloisite). 30 cm × 20 cm (Fig. 1a) were prepared using a thermal heat sealer Polikrimper TX/08 (Alipack, Pontecurone, Italy), provided by smooth bars at 130 °C for 0.5 s and 4.5 bar pressure. PVC (11 µm thick) stretchable film and EPS trays (Fungorobica srl, Cenate Sotto, Italy) were used as a control packaging configuration (Fig. 1b). Permeability properties of both materials against oxygen, carbon dioxide, and water vapor, expressed as oxygen transmission rate (OTR), carbon dioxide transmission rate (CO2TR), and water vapor transmission rate (WVTR), respectively, are reported in Table S1 of Supporting information.

Chitosan powder from crab shells (degree of deacetylation: 75–85%; molar mass distribution: 190,000–310,000; viscosity range: 200–800 cP, 1 wt.% in 1% acetic acid at 25 $^{\circ}$ C by Brookfield method) was purchased from Sigma Aldrich (Milano, Italy) and used without further purification.

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