

## THE INFLUENCE OF PACKAGING AND STORAGE TEMPERATURE ON THE CHEMICAL COMPOSITION OF FRESH OYSTER MUSHROOMS (*PLEUROTUS SAJOR-CAJU*) DURING STORAGE

Nguyen Thi Ngoc Giang<sup>1✉</sup>, Tran Van Khai<sup>2</sup>, Nguyen Minh Thuy<sup>3</sup>

<sup>1</sup>Experimental-Practical Area, An Giang University, Vietnam National University Ho Chi Minh city  
Ung Van Khiem 18, Long Xuyen city, An Giang province, **Vietnam**

<sup>2</sup>Crop Science Department, An Giang University, Vietnam National University Ho Chi Minh city  
Ung Van Khiem 18, Long Xuyen city, An Giang province, **Vietnam**

<sup>3</sup>Department of Food Technology, College of Agriculture, Can Tho University  
3/2 street, Can Tho city, **Vietnam**

### ABSTRACT

**Background.** *Pleurotus sajor-caju* has nutritional and medicinal values but it cannot be stored for more than 24 hours at room temperature because of its high water content. In addition, using the Modified Atmosphere in Packaging (MAP) method to store fresh mushrooms is an effective way to extend their shelf life.

**Materials and methods.** The protein, total sugar and lipid contents were determined for fresh oyster mushrooms packed in paper (P), high-density polyethylene (HDPE) and polyethylene terephthalate (PET) at 3–5°C and 28–30°C (with 60–62% and 76–78% of air humidity, respectively) during storage. The kinetics of chemical degradation were explored, and the best model was selected based on the highest  $R^2$  value and the lowest RMSE value.

**Results.** The loss of protein, total sugar and total lipid contents were lowest in the samples packed in HDPE packaging. Changes in the kinetics of protein and total sugar in different packaging and temperatures during storage followed the first-order model, while changes in total lipid followed the zero-order model. The half-life ( $t_{1/2}$ ) values of the samples at 3–5°C were 4–5 times higher than at 28–30°C.

**Conclusion.** The use of HDPE packaging was effective in maintaining the composition of *Pleurotus sajor-caju* at 28–30°C for 2–3 days and 13–15 days at 3–5°C.

**Keywords:** chemical composition, kinetic, oyster mushrooms, packaging, temperature, storage

### INTRODUCTION

*Pleurotus* spp. is an edible mushroom which is the most successfully grown and commercially important abalone mushroom (Zang et al., 2002). *Pleurotus sajor-caju* has nutritional and medicinal value. Oyster mushrooms have carbohydrates and proteins as the main components, accounting for 70 to 90% of the

dry weight, which are considered to be polymers with biological activity that protects human gut microbiota (Jayachandran et al., 2017). However, due to the high water content of oyster mushrooms (about 90%), they are susceptible to spoilage by microorganisms and reactions that depend on water content (Ares et al.,

✉ntngiang@agu.edu.vn, <https://orcid.org/0000-0002-2159-7278>

2007). Therefore, oyster mushrooms can be stored for barely more than 24 hours at room temperature (Satish et al., 2006). After harvest, mushrooms lose weight, turn brown, wilt and spoil. These reactions are mainly caused by respiration and transpiration.

Packaging, temperature and storage time greatly affect the quality of fresh oyster mushrooms (Silvia et al., 2014). Many previous studies have investigated the effects of packaging and temperature on the shelf life of oyster mushrooms. Fresh oyster mushrooms have been packed in low-density polyethylene (LDPE) bags and stored at 6–8°C for 6 days (Illeperuma and Jayathunge, 2004; Xiao et al., 2011), and in Polypropylene (PP) bags (thickness: 75 µm) for 15 days at 3°C (Rahman et al., 2021). The Modified Atmosphere in Packaging (MAP) method is effective in reducing the moisture loss of mushrooms because of the low water transfer rate of the packaging. Water vapor saturation and changes in the oxygen and CO<sub>2</sub> ratio inside the packaging help the mushrooms to reduce weight loss and spoilage and to maintain quality during postharvest (Antmann et al., 2008). The Modified Atmosphere in Packaging is a very effective and economical method for extending the shelf life of fresh mushrooms during transportation and marketing. However, excessive accumulation of CO<sub>2</sub> in the packaging causes deterioration of firmness and brown development due to cell damage (Kader, 2002). In addition, oyster mushrooms have high metabolic and respiration rates compared to vegetables and fruits (Mahajan et al., 2008; Iqbal et al., 2009). The rate of respiration is inversely proportional to the storage time. Several factors affect the respiration rate, the most important of which is temperature, to help mushrooms maintain quality during storage (Azevedo et al., 2017). Storage temperature has a strong influence on the overall physiological and biochemical processes and microbial activities (Silvia et al., 2014). The objective of this study was to implement the appropriate packaging and temperature parameters to prolong the shelf life of *Pleurotus sajor-caju* and to determine the changes in chemical composition during storage.

## MATERIALS AND METHODS

### Materials

*Pleurotus sajor-caju* mushrooms were selected by An Giang University, Vietnam University Ho Chi Minh

city (Vietnam) according to Singh's standards (2011; 12.5 ±2.5 cm in length, 6.6 ±3.5 cm cap diameter and 10.48 ±4.32 g in weight). The fresh mushrooms were washed with water before being soaked for 10 minutes in 1.5% CaCl<sub>2</sub> and 15 minutes in 1% citric acid to improve the color and firmness during storage.

Three commercial packaging materials were used: paper (P; thickness: 83.33 µm), high-density polyethylene (HDPE; thickness: 91.72 µm) and polyethylene terephthalate (PET; thickness: 250.33 µm). The size of the packaging was 27×20.5 cm for all materials. The O<sub>2</sub> and CO<sub>2</sub> of the paper packaging were undetermined, while those of HDPE were 16 and 72 and those of PET were 0.27–0.39 and 5.1 cc-mm/m<sup>2</sup>-24 hr.-Bar, respectively (Mark and John, 2003).

### Experimental design

100 g of soaked mushrooms were packed and stored at 3–5°C and 28–30°C (with air humidity of 60–62% and 76–78%, respectively). The samples were made every 2 days (for samples at 3–5°C) and every day (for samples at 28–30°C) and analyzed for changes in chemical composition. The number of samples taken for analysis was 3 for each treatment.

### Determination of protein

The total protein content (g/100 g dry matter) was measured using the Lowry method (Nielsen, 2010) with some modifications. 1 g of fresh sample and 10 mL of concentrated sulfuric acid were added to a Kjeldahl tube and digested on a digestion block until the solution of the solvent was clear. 0.1 mL of digested sample or standard was combined with 0.1 mL of 2 N NaOH in the tube and placed in boiling water for 10 minutes. The tube was cooled to room temperature and then 1 mL of complex forming reagent was added (including 2% (w/v) Na<sub>2</sub>CO<sub>3</sub>, 1% (w/v) CuSO<sub>4</sub>·5H<sub>2</sub>O and 2% (w/v) sodium potassium tartrate in the proportion of 100:1:1, respectively). Approximately 0.1 mL of Folin-Ciocalteu reagent was then added into the tube, which was then vortexed and left at room temperature for 30 minutes. The Lowry method is based on the reaction of Cu<sup>+</sup> with Folin-Ciocalteu reagent to react into an intense blue colour and measured with an absorbance of 750 nm. The concentration of total protein was based on the standard curve of protein:

$$y = 0.0041x + 0.0118 \quad (R^2 = 0.9999)$$

where:

- $y$  – the absorbance,
- $x$  – the concentration of the solution in the tube.

### Determination of total sugar

Total sugar (g/100 g dry matter) was measured using the DNS method (Nielsen, 2010) with some modifications. This method is based on the oxidation of the C = O group by 3,5-dinitrosalicylic acid from a yellow colour to orange red in an alkaline medium. 1 g of fresh sample and 5 mL of concentrated HCl were added to 100 mL and placed in a boiling water bath for 30 minutes. Protein and impurities were precipitated with 30% (w/v)  $\text{Pb}(\text{C}_2\text{H}_3\text{O}_2)_2 \cdot 3\text{H}_2\text{O}$ , 18–20 mL of saturated  $\text{Na}_2\text{SO}_4$  solution was added (to remove excess  $\text{Pb}(\text{C}_2\text{H}_3\text{O}_2)_2 \cdot 3\text{H}_2\text{O}$ ), and the mixture was filled up to the mark with distilled water and filtered through filter paper. An aliquot (1 mL) of solution was put in a test tube and then 2 mL of reagent DNS was added. All of the tubes, blank, the solution of standard glucose and the samples, were put in boiling water for 10 minutes. Next, 7 mL of distilled water was added. The solution was analysed at an absorption of 575 nm. The concentration of total sugar was based on a standard curve of glucose:

$$y = 23885x + 0.126 \quad (R^2 = 0.9999)$$

where:

- $y$  – the absorbance,
- $x$  – the concentration of the solution in the tube.

### Determination of total lipid

Total lipid was determined using the Soxhlet method (Nielsen, 2010). The samples were dried at 105°C to a constant weight. 5 g of dried sample was put in the thimble and 350 mL petroleum ether in the flask of a Soxhlet extractor and extracted for 24 hours. After the end of the extract, the sample was removed from the thimble, the solvent was evaporated and the sample was dried to a constant weight. The lipid content was measured according to equation 1.

$$X = \frac{(a - b) \cdot 100}{a} \quad (1)$$

where:

- $X$  – lipid content, %,
- $a$  – weight of dried sample before extraction, g,
- $b$  – weight of sample after extraction, g.

### Computation of reaction kinetics

Zero, first and second order kinetic models were calculated according to equations 2, 3 and 4. The best model for each function was selected based on the highest  $R^2$  value and the lowest root mean squared error (RMSE) value (Charurungsipong et al., 2020).

$$\text{Zero-order:} \quad C = -kt + C_0 \quad (2)$$

$$\text{First-order:} \quad \ln\left(\frac{C}{C_0}\right) = -kt \quad (3)$$

$$\text{Second-order:} \quad \frac{1}{C} - \frac{1}{C_0} = kt \quad (4)$$

The half-life ( $t_{1/2}$ ) was calculated according to equation 5.

$$t_{1/2} = \frac{\ln 2}{k} \quad (5)$$

where:

- $C_0$  – an initial target function (protein, total sugar or lipid) content, g/100 g of dry matter,
- $C$  – a target function content after time of storage, day,
- $k$  – the reaction constant of the rate of target function degradation, day<sup>-1</sup>,
- $t_{1/2}$  – the half-life of the target function, day.

### Data analysis methods

Data were collected and processed using Microsoft Excel software for calculating and graphing.

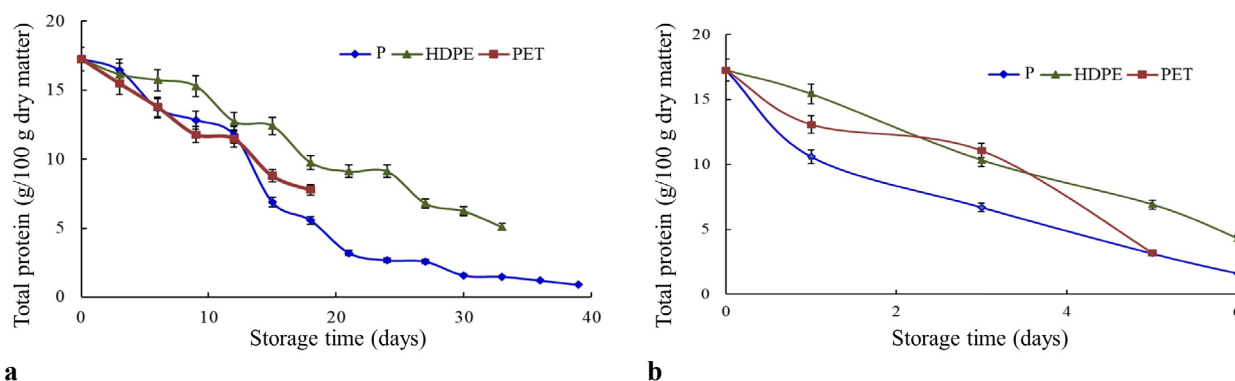
## RESULTS AND DISCUSSION

### Changes in chemical composition of *Pleurotus sajor-caju* at different packagings and temperatures during storage

Soaked oyster mushrooms packed in P, HDPE and PET and stored at 3–5°C for 39, 33 and 18 days, respectively, and at 28–30°C for 5 to 6 days.

**Changes in protein content during storage.** The effects of packaging and storage temperature on the change in protein content are shown in Figure 1.

The results showed that the packaging and storage temperature had different effects on the protein content of fresh oyster mushrooms. The samples stored at



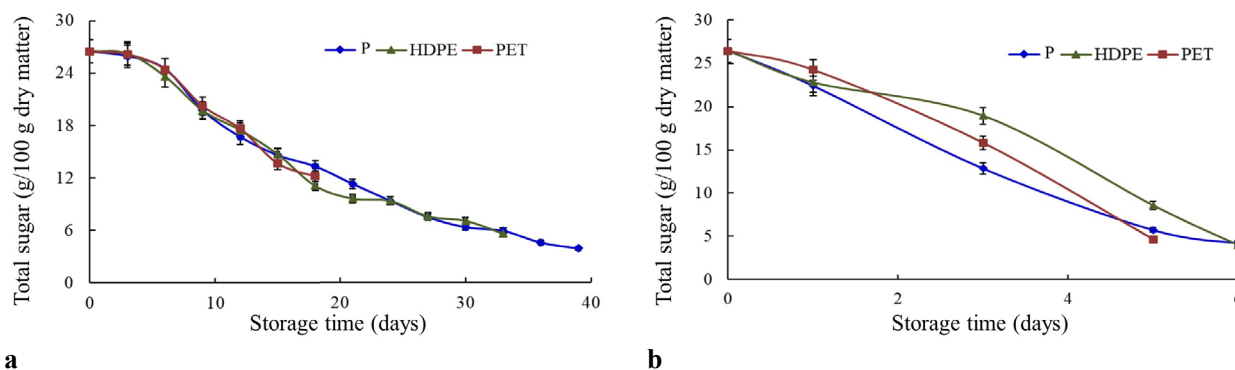
**Fig. 1.** Changes in protein content during storage: **a** – 3–5°C, **b** – 28–30°C

28–30°C lost more protein content than the samples at 3–5°C. The protein content started to decrease rapidly after 3 days at 28–30°C and after 12 days at 3–5°C. The losses of protein in P, HDPE and PET were 61.28–81.90%, 40.14–59.95% and 35.91–81.42%, respectively, at 28–30°C after 3–5 days of storage. At 3–5°C, the protein losses were 31.87–60.16%, 26.1–28.12% and 33.61–49.06% after 12–15 days in the P, HDPE and PET packaging, respectively. The reduction in protein content or protein oxidation is influenced by many factors, including the activity of tyrosinase (Seo et al., 2003). In addition, the protein of *Pleurotus sajor-caju* contains amino acids, such as cysteine, lysine, histidine, methionine and tryptophane, that are easily oxidized (Kayode et al., 2015). Oxidative reactions can lead to protein degradation and loss of function (Xiong and Guo, 2021). Besides this, protein

oxidation still took place during cold storage (Hambly and Gross, 2009). The decrease in protein content was also related to non-enzyme browning (Maillard reaction) between amino acids and reducing sugars.

**Changes in total sugar during storage.** The changes in total sugar content with storage time in different packaging and storage temperatures are shown in Figure 2.

The total sugar loss was lowest in the HDPE packaging at 28–30°C after 3–4 days and at 3–5°C after 12–15 days (28.45–40.25% and 33.24–44.47%, respectively). During storage, mannitol and  $\alpha$ -trehalose, which are the main components of the polysaccharides in oyster mushrooms, decreased, and the total sugar content also decreased (Reis et al., 2012; Kalac, 2012). Oyster mushrooms have higher metabolic and



**Fig. 2.** Changes in total sugar content during storage: **a** – 3–5°C, **b** – 28–30°C

respiration rates than vegetables and fruits (Mahajan et al., 2008; Iqbal et al., 2009). The carbon content of mushrooms is reduced as a result of CO<sub>2</sub> production by respiration, even at low temperatures (Singh et al., 2011). Fresh products are often stored in semi-permeable packaging (such as polyethylene (PE) and polyethylene terephthalate (PET)) because of their continuing respiration, which leads to changes in the gas composition of the packaging. After this, when combined with low temperatures, it reduces the rate of respiration and microbial growth and thus prolongs shelf life (Barbosa et al., 2011).

In addition, during storage, the Maillard browning reaction is also activated, even at 0°C, and with every 10°C increase in temperature, the reaction speed increases 2–3 times (Verma et al., 2019). *Pleurotus sajor-caju* contains sugars and amino acids, so browning is inevitable when mushrooms are stored at temperatures higher than 5°C.

**Changes in total lipid during storage.** The changes in total lipid content with storage time in different packagings and storage temperatures are shown in Figure 3.

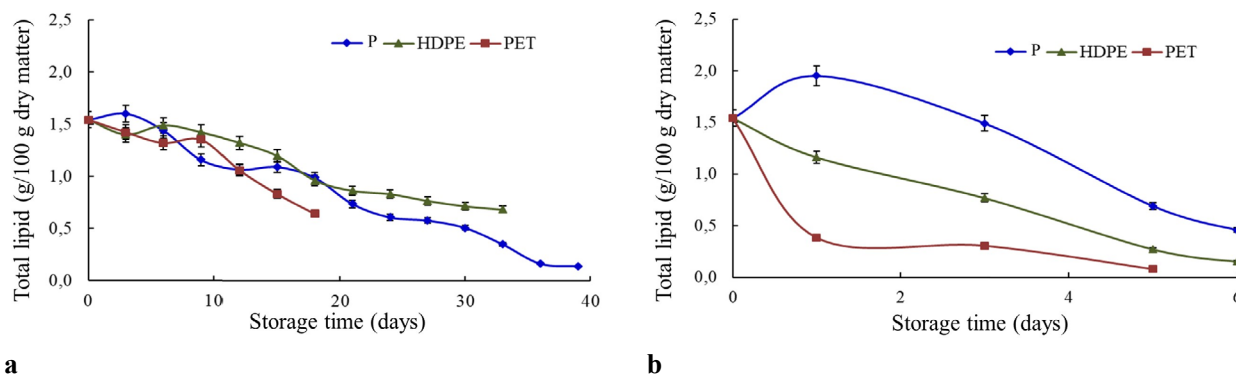
The total lipid content of fresh oyster mushrooms when stored in P at 28–30°C increased after 1 day of storage and decreased rapidly with further increases in storage time (Fig. 3b). In contrast, in the fresh oyster mushroom samples stored at 3–5°C, the total lipid content decreased with time of storage (Fig. 3a). The total lipid content underwent its lowest decrease in the

P packaging after 5 days at 28–30°C and in the HDPE packaging after 12–15 days at 3–5°C (55.19% and 14.15–22.30%, respectively). During the storage of fresh oyster mushrooms, the protein is oxidized to free amino acids, which are further oxidized and converted to acetyl coenzymes. Fatty acids are synthesized by acetyl coenzymes (Rai and Arumuganathan, 2008). Research by Ambatkar (2012) also found an increase in the fat content of samples with different packagings and temperatures. Lower temperatures reduce the intensity of oxidation, but it does not stop due to the formation of free radicals during oxidation, which is stable at low temperatures (Liu et al., 2019).

#### Kinetics of chemical degradation using different packaging materials and temperatures during storage

The kinetic models (with zero, first and second order models) of protein degradation in different packaging and storage temperatures with storage time are shown in Table 1. It was found that the degradation of protein followed the first-order model because of the highest R<sup>2</sup> and smallest RMSE. This result is in agreement with the research of Ovissonpour et al. (2017) on the changes in protein of *Salmo salar* during storage time.

The kinetic models of protein and lipid degradation in different packaging and storage temperatures with storage time are shown in Tables 2 and 3. Similar to protein degradation, the results show that degradation of total sugar followed the first-order reaction, which was also found by Okoro and Odeunmi (2009). In



**Fig. 3.** Changes in total lipid content during storage: **a** – 3–5°C, **b** – 28–30°C

**Table 1.** The kinetic models of protein degradation in different packaging and storage temperatures

Packaging	Temperature, °C					
	28–30			3–5		
	equation	$R^2$	RMSE	equation	$R^2$	RMSE
Zero-order model: $C = -kt + C_0$						
P	$y = -2.150x + 17.301$	0.998	0.312	$y = -0.461x + 16.001$	0.952	0.741
HDPE	$y = -2.382x + 14.998$	0.964	1.509	$y = -0.385x + 17.659$	0.990	0.131
PET	$y = -2.581x + 16.960$	0.971	1.423	$y = -0.526x + 15.297$	0.993	0.080
First-order model: $\ln(C/C_0) = -kt$						
P	$y = -0.369x$	0.983	0.029	$y = -0.037x$	0.982	0.157
HDPE	$y = -0.222x$	0.988	0.105	$y = -0.083x$	0.979	0.024
PET	$y = -0.313x$	0.932	0.153	$y = -0.044x$	0.988	0.008
Second-order model: $(1/C) - (1/C_0) = kt$						
P	$y = 0.083x$	0.932	0.124	$y = 0.025x$	0.952	0.160
HDPE	$y = 0.023x$	0.906	0.131	$y = 0.004x$	0.990	0.080
PET	$y = 0.047x$	0.882	0.255	$y = 0.004x$	0.993	0.164

$y$  – protein content, g/100 g of dry matter,  $x$  – storage time, days.

**Table 2.** The kinetic models of total sugar degradation in different packaging and storage temperatures

Packaging	Temperature, °C					
	28–30			3–5		
	equation	$R^2$	RMSE	equation	$R^2$	RMSE
Zero-order model: $C = -kt + C_0$						
P	$y = 3.851x + 25.894$	0.993	1.060	$y = 0.625x + 25.776$	0.980	0.975
HDPE	$y = 3.694x + 27.237$	0.986	1.427	$y = 0.692x + 26.291$	0.978	0.747
PET	$y = 4.425x + 27.764$	0.991	1.298	$y = 0.888x + 26.137$	0.982	2.059
First-order model: $\ln(C/C_0) = -kt$						
P	$y = -0.317x$	0.994	0.022	$y = -0.050x$	0.995	0.022
HDPE	$y = -0.294x$	0.941	0.042	$y = -0.040x$	0.993	0.011
PET	$y = -0.343x$	0.949	0.033	$y = -0.051x$	0.973	0.009
Second-order model: $(1/C) - (1/C_0) = kt$						
P	$y = 0.033x$	0.960	1.550	$y = 0.005x$	0.980	0.064
HDPE	$y = 0.030x$	0.856	0.242	$y = 0.004x$	0.978	0.063
PET	$y = 0.034x$	0.893	0.219	$y = 0.003x$	0.982	1.577

$y$  – total sugar content, g/100 g of dry matter,  $x$  – storage time, days.

**Table 3.** The kinetic models of lipid degradation in different packaging and storage temperatures

Packaging	Temperature, °C					
	28–30			3–5		
	equation	$R^2$	RMSE	equation	$R^2$	RMSE
Zero-order model: $C = -kt + C_0$						
P	$y = 0.222x + 1.894$	0.912	0.242	$y = 0.038x + 1.607$	0.990	0.510
HDPE	$y = 0.229x + 1.467$	0.994	0.059	$y = 0.029x + 1.584$	0.971	0.055
PET	$y = 0.237x + 1.113$	0.951	0.342	$y = 0.049x + 1.851$	0.962	0.199
First-order model: $\ln(C/C_0) = -kt$						
P	$y = -0.220x$	0.911	0.319	$y = -0.058x$	0.945	0.252
HDPE	$y = -0.378x$	0.981	0.170	$y = -0.028x$	0.974	0.067
PET	$y = -0.516x$	0.945	0.374	$y = -0.046x$	0.939	1.574
Second-order model: $(1/C) - (1/C_0) = kt$						
P	$y = 0.249x$	0.895	0.289	$y = 0.129x$	0.990	1.164
HDPE	$y = 0.895x$	0.929	0.929	$y = 0.027x$	0.971	0.070
PET	$y = 2.177x$	0.803	1.860	$y = 0.046x$	0.962	0.226

$y$  – lipid content, g/100 g of dry matter,  $x$  – storage time, days.

**Table 4.** The half-life ( $t_{1/2}$ ) of protein, total sugar and lipids in different packaging and storage temperatures

Packaging	$t_{1/2}$ , day					
	protein		total sugar		lipid	
	28–30°C	3–5°C	28–30°C	3–5°C	28–30°C	3–5°C
P	1.879	8.372	2.190	13.949	3.121	18.027
HDPE	3.122	18.698	2.353	14.741	3.029	23.657
PET	2.215	15.696	2.020	13.483	2.920	14.157

contrast, changes in the total lipid content followed the zero-order model, which is similar to a study by Husain et al. (2016).

The half-life ( $t_{1/2}$ ) values of protein, total sugar and lipids in different packaging and storage temperatures are shown in Table 4. The results indicated that the half-life ( $t_{1/2}$ ) values of the samples at 3–5°C were 4–5 times higher than at 28–30°C. Besides this, the half-life of the samples in HDPE packaging was higher than the others, reaching 3.122 and 14.741 when

storing oyster mushrooms at 28–30°C and 3–5°C, respectively. The shelf life at 3–5°C was 4–5 times longer than at 28–30°C.

## CONCLUSION

Packaging and storage temperature have great effects on changes in the chemical composition of oyster mushrooms. The half-life of the mushrooms in HDPE packaging was higher than that of P and

PET. Therefore, the use of HDPE packaging is recommended in order to minimize the loss of nutrients from oyster mushrooms during storage. The fresh oyster mushrooms were packed in HDPE and stored at 28–30°C for 2–3 days and 13–15 days at 3–5°C.

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