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# Influence of canning process on colour, weight and grade of mushrooms

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## Abstract

According to Spanish legislation, some of the quality parameters of canned mushroom (colour, weight and grade) have been studied. Different times of blanching treatment and two brines, with and without ascorbic acid, were used. Blanching had an important effect on the final state of mushrooms, decreasing the losses of weight and grade and improving the colour, which was clear and pleasant. A positive effect of ascorbic acid was observed. The presence of this acid in the brine improves colour stability and acceptance. It inhibited the browning process. The pH of brine plus mushroom is independent of brine composition (with or without ascorbic acid). Longer times of blanching had no significant effects on mushroom quality parameters.  $\odot$  1999 Elsevier Science Ltd. All rights reserved.

## 1. Introduction

Canning of mushroom induces important changes in its physical and chemical properties. One of the most remarkable and important changes is the loss of water from the mushroom, which produces a loss of weight and a reduction of the net weight in the can. These losses have an important economic repercussion (Coale & Burtz, 1972; Gormley & McCanna, 1979; Maggioni & Renosto, 1970).

The colour changes, which modify the acceptance of canned mushrooms, are also important. The mushroom browns very quickly during the canning process, due to the action of polyphenoloxidase (PPO), also called diphenoloxidase or tyrosinase. The main substrate for PPO activity in mushrooms is the tyrosine. The enzyme transforms tyrosine into 3,4-dihydroxyphenylalanine (L-DOPA), and afterwards, into DOPAquinone. From this compound, several reactions which take place spontaneously, originate the melanins (brown pigments) (García, Cabanes, & Garcia-Canovas, 1987; Jiménez et al., 1984). Some of the most important factors that contribute to the enzymatic browning of vegetables are the levels of phenolic compounds and PPO activity, the pH, the temperature and the oxygen available in the tissue (Martínez & Whitaker, 1995).

The aim of the present work was to study effects of the mushroom canning process with special attention to the effects of the blanching process, and the composition of brine on colour, weight and final grade of mushrooms. These are parameters of mushroom quality according to the Spanish legislation on Technical-Sanitary regulation for Elaboration and Commercialisation of Canned Vegetables', adapted to the E.U. legislation.

## 2. Materials and methods

## 2.1. Raw material

Commercial fresh mushrooms (Agaricus bisporus) were canned in small-scale food processing equipment of the Biotechnology and Food Science Department of the University of Burgos. The mushrooms were bought in the market on the same day that they were processed.

## 2.2. Canning process

Fig. 1 shows the canning process applied in this study. This figure shows the different blanching treatments and brines used. At the end, six different lots of cans were studied, three with the brine 1 and three with the brine 2 (Table 1). For each of the six lots, 30 cans were elaborated.

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Table 1 Lots investigated and codex used for their identification

Blanching treatment	<b>Brine</b>	Codex	
Without	1 2\% NaCl + $0.5\%$ citric acid Control B1		
	2 $2\%$ NaCl + 0.5% citric acid Control B2		
	$+1000$ ppm ascorbic acid		
8 min	1 2\% NaCl + $0.5\%$ citric acid	8t B1	
95 $\rm ^{\circ}C$ (H <sub>2</sub> O)	2 2\% NaCl + $0.5\%$ citric acid	8t B <sub>2</sub>	
	$+1000$ ppm ascorbic acid		
$10 \text{ min}$	1 2\% NaCl + $0.5\%$ citric acid	$10t$ B <sub>1</sub>	
$95^{\circ}$ C (H <sub>2</sub> O)	2 2% NaCl + $0.5%$ citric acid	$10 \text{ t}$ B <sub>2</sub>	
	$+1000$ ppm ascorbic acid		

Two brines were used in order to confirm the effect of the ascorbic acid on mushroom quality; this acid has been described as a good antioxidant for control of browning of foods (Sapers & Miller, 1993).

The cans were stored for three months at room temperature ( $\pm 20^{\circ}$ C).

#### 2.3. Determination of weight and grade

Weight was determined, according to current guidelines, by weighting the product after draining the can content for two min in a perforated stainless steel tray. The determination of grade was done by measuring the diameter of the three caps per can with a Vernier caliper.

Both parameters were determined after blanching and cooling (sample 1 in Fig. 1) and when the canning was completely elaborated (sample 2 in Fig. 1). The weight and grade losses during the process of sterilisation were calculated and expressed as percentage of lost weight or grade.

## 2.4. Determination of pH and colour

The pH of the brine and the mushroom was measured using a Micro pHmeter 2000 of Crison.

The colour was determined by using a range of colours made for that purpose. In this way, a range series of colours was codified with alphanumeric codex (Table 2).

The colour and its acceptance were evaluated using 10 tasters who were not trained but who were usual consumers of canned mushrooms. These tasters were asked to point out which mushrooms had good colour, and which ones presented bad or defective colours.

## 2.5. Determination of phenolic content and PPO activity

PPO activity and phenolic total contents were measured. These parameters are principally responsible for the colour change of the mushrooms.



Fig. 1. Mushroom canning process.

#### 2.5.1. Total phenolic content

Ten grams of mushrooms were homogenised with 20 ml of methanol:formic acid (95:5), using a Ultraturrax T-25.

The homogenate remained for 24 h at  $-20^{\circ}$ C then the liquid part was separated by filtering. The solid residue was extracted twice in the same way. The three liquid extracts were combined and concentrated in a vacuum evaporator down to more or less 25 ml. In this process, the temperature was less than  $35^{\circ}$ C. Distilled water was added in order to obtain a final volume of 50 ml.

Two different phenolic families were determined in this extract: total polyphenols (TP) and orthodiphenols

Table 2 Alphanumeric codex used for colour and evolution of canned mushrooms colour during storage

Blanching treatment		Storage (months)					
	<b>Brine</b>	$\mathbf{0}$	0.5		1.5		3
Control		4B <sup>a</sup>	4B	4B	4C	4C	4C
		4A	3A	4A	4A	4A	4A
8t		2A	3A	4B	4C	4C	4C
		3A	4A	4A	4A	4A	4A
10 <sub>t</sub>		3A	2A	4C	4C	4C	4C
		2A	4A	3A	4A	4A	4A

<sup>a</sup> Colour: A, cream B, yellow C, brown D, dun. Tonality: 1 to 5 notes the darkness of colour. (1C=Clear brown as honey; 5C=dark brown as chocolate.

(OD). Classic methods were used, based on the capacity for reaction with the Arnow reactive (OD) and with the Folin±Ciocalteu reactive species (TP) (Paronetto, 1977).

Four measures were performed for each blanching treatment and brine.

# 2.5.2. Extraction of PPO and determination of PPO activity

Ten grams of mushrooms were homogenised with 20 ml of ice-cold acetone  $(-20^{\circ}C)$  using an Ultraturrax T-25. The homogenate obtained was filtered and the solid residue was washed with 20 ml of cold acetone, twice. Then the solid extract was dried in a vacuum evaporating dish, where it remained for 24 h at a controlled temperature  $(10^{\circ}$ C). This extract is stable and can be kept refrigerated for at least two months, without suffering loss of PPO activity.

The determination of PPO activity was performed on a liquid extract obtained from the solid extract (Ngalani, Signoret, & Crouzet, 1993). The solid extract (0.03 g) was suspended in 5 ml of the pH 6.5 McIlvain buffer (Na<sub>2</sub>HPO<sub>4</sub>, 0.1 M, adjusting pH with citric acid). This suspension was stirred with cold water  $(4^{\circ}C)$  and in darkness for 20 min. After that period, suspension was filtered through a Whatman No. 1 filter paper, which had been previously wet with the cold buffer. The filtered extract is unstable and must be used immediately.

PPO activity was measured according to Coseteng and Lee (1987). A solution of gallic acid  $(1.5 \text{ g/l})$  made in an acetate buffer at 5.8 pH  $(1 \text{ ml})$ , distilled water  $(0.5 \text{ ml})$  and 1 ml of the obtained extract were put in a 1 cm pathlength glass cuvette. Then the increase of absorbance at 420 nm during 1 h was measured. A spectrophotometer (Beckman DU-650) was used. The measurements were made at 25C. Four measurements were performed for each treatment, brine and storage time.

Under these conditions, PPO activity was determined as the amount of brown polymers formed from a known concentration of gallic acid, quantified as the increase in absorbance registered at 420 nm per unit of time.

All of these parameters were measured at the end of the canning process and every two weeks, using five cans every time.

# 2.6. Statistical analysis

In order to determine statistically significant differences among treatments, an ANOVA (analysis of variance) was applied by STATGRAPHICS PLUS<sup>R</sup> software.

## 3. Results and discussion

## 3.1. Weight

No brine effects were detected for any blanching treatment. That is, there were no statistically significant differences between the weight losses of each brine lot (Fig. 2).

The highest losses of weight took place in the control treatments, and they were statistically different from the losses of blanched mushrooms.

These data show the important effect of blanching. When mushrooms were not blanched, their final weight losses were at least twice that of blanched mushroom.





Fig. 2. Percentage loss in net weight due to canning process. Two similar letters mean that there are no statistically significant differences between values.

In addition, the data showed that it is necessary to know the percentage of loss in order to control the net weight, which must be shown on the label of the cans. If the losses are not evaluated, some mistakes in production could be made, causing important economic repercussions for the company. On the one hand, if the losses are not correctly taken into account, net weights inferior to the expected ones will be obtained, resulting in fraud and a reason for sanctions. On the other hand, when losses are lower the net weight would be superior to the expected one, which is not punishable, but implies economic losses, because the product is being sold at a lower price than that corresponding to its real content.

## 3.2. Grade

The data relating to the grade losses (Fig. 3) showed similar results to weight losses. A brine effect was not detected. However, in this case there were not only statistically significant differences between blanched and unblanched mushrooms, but also between the two treatments of blanching used.

The lowest losses of grade were detected for the longest treatment of blanching (10 min), which were four times inferior to the detected losses on non-blanched mushrooms.

These and previous results (weight losses) revealed that a higher loss of grade does not always correspond



Fig. 3. Percentage reduction in the grade of canned mushrooms due to canning process. Two similar letters mean that there are no statistically significant differences between values.



with a higher loss of weight. This has an important repercussion in mushroom commercial quality. The classification in quality of commercial class relates to the grade of final product, lower level of quality corresponding to a lower grade of mushroom.

## 3.3. pH

No brine or blanching effect on pH of mushroom or brine was detected (Table 3), presumably due to a buffering capacity effect.

## 3.4. Phenolic compounds, PPO activity and colour

Table 4 shows that the canning process produced a significant loss of the mushroom total phenolic content (TP). These losses were higher when brine 1 was used and the losses were independent of the type of blanching treatment.

Phenolic compounds are thermally unstable, have a high tendency to oxidation, polymerisation and condensation, and are soluble in water (element in which mushroom is immersed during all the processes). All of these explain the observed losses. The presence of ascorbic acid, an antioxidant, in brine 2, justifies the lower losses of TP in this brine.

PPO activity showed a drastic reduction in canned mushroom. This reduction was due to various factors:

Total polyphenolic (TP) content, orthodiphenols (OD), and PPO activity in canned and fresh mushrooms. Two numbers followed by the same letter do not differ significantly





<sup>a</sup> Storage (months).

Table 4

- 1. the treatment with metabisulfite which plays an inhibiting role on PPO (Osuga & Whitaker, 1996);
- 2. the thermal treatment, which originates the inactivation of the enzyme (Martínez  $\&$  Whitaker, 1995);
- 3. auto-inactivation. The PPO activity decreases when the enzyme acts (Bajaj, Diez, Junquera, & González-Sanjosé, 1997).

The statistical results show that reduction of PPO activity is independent of the applied blanching treatment. The effect of blanching process on PPO activity is minimised, due to the thermal inactivation of PPO that occurs principally during the sterilisation process.

Statistical difference was found between PPO activity of two treatments: control B2 and 8t-B2, the other four treatments, being lower. These are surprising results. According to previous comments, higher PPO activity in B2 canned mushrooms was expected.

Orthodiphenols are very unstable compounds, as expected, they were not found in canned mushroom.

The colour of canned mushroom presented differences between applied blanching treatments (Table 2). Since control mushrooms were evaluated with darker colours, a positive effect of blanching process on the mushroom colour is indicated. The tasters also reported that the mushrooms elaborated with brine 2 had better colours, and the presence of ascorbic acid in brine 2 improved the mushroom colour. Luo and Barbosa-Cánovas (1995) noted similar effects of ascorbic acid on apple colour.

The obtained results of colour and phenolic compounds are closely correlated. It was observed that the darkest mushrooms had the lowest phenolic content, which is due to a greater browning process in which the phenolic compounds are involved. However, under unfavourable conditions for browning, e.g. the presence of ascorbic acid, high levels of total phenolics remained in mushrooms, and their colours were clearer because the phenolic compounds were not transformed into brown pigments.

During the first three months of storage, evolution of mushroom colour (Table 2), total polyphenol content and residual PPO activity (Figs. 4 and 5), and pH of brine and mushroom (Table 3) were studied.

Mushrooms and brine pHs remained constant without differences between samples. Total polyphenol levels decreased slowly, with the exception of Control B2, in which a significant decrease of phenolic compounds is detected. PPO activity decreased significantly during the first two months and afterwards it did it more slowly, independently of the brine composition. A momentary reactivation of residual PPO could be the reason for the observed increase in PPO activity after 0.5 month's storage, in some of the treatments.

Colours of mushrooms became darker with the storage time. In cans elaborated with brine 1, changes of colour from cream to brown were observed. However,



Fig. 4. Evolution of total polyphenols (TP) content during the storage of the cans.



Fig. 5. Evolution of PPO activity during the storage of the cans.

in canned mushroom elaborated with brine 2 only tonality increases were detected. In addition, the tasters noted that canned mushrooms elaborated with brine 1 were very dark, their colour was unpleasant and a lot of them showed superficial black spots.

In conclusion, the use of ascorbic acid in the brine improved the colour and acceptance of canned mushrooms and did not modify pH, therefore the use of this acid is recommended.

On the other hand, the blanching process reduced the losses of weight and grade of mushrooms, improved their colour and did not modify the pH.

However, more intensive blanching processes (more time) did not produce significant improvement of the quality parameters; long times for this process are not recommended.

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