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Journal of Food Engineering 77 (2006) 1078–1086
JOURNAL OF FOOD ENGINEERING
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# Influence of modified atmosphere packaging on the chilled shelf life of gutted farmed bass (*Dicentrarchus labrax*)

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Received 6 June 2005; accepted 2 August 2005
Available online 28 September 2005

**Abstract**

The effect of MAP on quality changes of gutted farmed bass when stored at 3 °C were investigated for up to 9 days. Gutted farmed bass was packed with six different atmospheres (0%$O_2$–70%$CO_2$; 20%$O_2$–70%$CO_2$; 30%$O_2$–60%$CO_2$; 40%$O_2$–60%$CO_2$; 30%$O_2$–50%$CO_2$; 21%$O_2$–0%$CO_2$). Headspace gas composition ($O_2$%; $CO_2$%), *aerobic mesophilic bacteria* (AMB) and *Enterobacteriaceae*, pH, water loss, flesh moisture content, colour, stiffness, odour and eyes appearance were assessed by means of instrumental and sensory analysis after 0, 2, 5, 7 and 9 days of storage. Atmosphere composed of 30% of $O_2$ and 50% of $CO_2$ was the best one to preserve the quality of the gutted farmed bass. PCA was an effectively instrument to classify gutted bass samples on the bases of quality changes. The effect of the time was explained by the factor 1, whereas the fish were clearly classified along factor 2 in relation to storage atmosphere.


*Keywords:* Gutted farmed bass; Modified atmosphere packaging; Quality changes; Shelf life

## 1. Introduction

Sea bass (*Dicentrarchus labrax*) is one of the main marine fish species aqua-cultured in the Mediterranean countries (Kyrana & Lougovois, 2002). Due to its high nutritional quality and excellent sensory properties, sea bass is one of the preferred fish species by the Italian consumers (Boyd, Green, & LePors, 1992; Poli et al., 2001). After fishing, the fishes are stored in foam polystyrene box covered by grounded ice and delivered to the fish market. The shelf life of the sea bass stored in ice is of about 6–8 days (Gricorakis, Alexis, Gialamas, & Nikolopoulou, 2004; Kyrana & Lougovois, 2002; Paleologos, Savvaidis, & Kontominas, 2004; Poli et al., 2001). Usually the sea bass, as the most part of fish species, is sold as it is, i.e. without boning or scaling and gutting it. However, in the recent years the demand from consumers and restaurants of ready to cook fish with a 6–8 days shelf life is largely increased. Papadopoulos, Chouliara, Badeka, Savvadis, and Kontominas (2003) reported on the quality assessment of whole and evischerated aquacultured sea bass stored in ice. Results of their study indicate that the gutted bass has a shorter shelf life than ungutted bass, respectively equal to 8 and 13 days. This may be attributed either to cross-contamination of fish during gutting procedures or to the significantly higher fish flesh surface area exposed to environmental microbial contamination in the case of gutted fish. The most suitable packaging technology for such a convenient but perishable product could be the modified atmosphere packaging (MAP), which is even an effective system to preserve the quality and to maintain the hygienic, sanitary and sensory characteristics of perishable foods (Corbo et al., 2005; Devlieghere & Debevere, 2003; Pastoriza, Sampedro, Herrera, & Cabo, 1998; Reddy & Armstrog, 1992; Stammen, Gerdes, & Caporaso, 1990). MAP, in conjunction with refrigeration, has been shown positive effects on fish shelf life (Sivertsvik, Jeksrud,

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doi:10.1016/j.jfoodeng.2005.08.038

& Rosnes, 2002). Nevertheless, raw material with high degree of freshness is required for fresh fish packed in MAP (Bøknæs, Österberg, Nielsen, & Dalgraad, 2000). The deterioration of fresh fish is due to bacterial and enzymatic action and during spoilage, fish undergo to colour, flavour and texture changes (Boyd et al., 1992; Colby & Flick, 1993; Gram & Huss, 1996). Carbon dioxide atmosphere, with variable proportions in oxygen, can be considered effectively inhibitory on the total aerobic flora (Stammen et al., 1990). On the other side, MAP can affect the quality of the product, mainly owing to $CO_2$ dissolved in the muscle tissue, which is associated with an increase of carbonic acid (Sivertsvik, Rosnes, & Jeksrud, 2004). A greater loss of water-holding capacity of muscle protein occurs at lower pH values. Randell, Hattula, and Ahvenainen (1997) reported that the shelf life of rainbow trout and Baltic herring fillet packed in modified atmosphere package (35% $CO_2$) was limited by the excessive drip formed during storage, that on the other hand was lower when the fish was packed not in a modified atmosphere package. Masniyom, Benjakul, and Visessanguan (2005), by studying the collagen changes in refrigerated sea bass muscle treated with pyrophosphate (PP) and stored in modified atmosphere package, indicated that $CO_2$ at a concentration of 80%, especially in combination with PP treatment, effectively inhibited the degradation of fish collagen caused by endogenous enzymes as well as those from microorganisms. That induces to maintained firmness with less detachment of sea bass muscle during extended storage. A mixture of 30%$O_2$–40%$CO_2$–30%$N_2$ or 40–60% of $CO_2$ and 60% $N_2$ has been proposed, respectively, for low fat fish and for fatty fish (Cann, 1984; Pastoriza et al., 1998; Robertson, 1993; Woyewoda, Bligh, & Shaw, 1984). Debevere and Boskou (1996) proposed a mixture of 40%$O_2$–60%$CO_2$ as the most effective for the inhibition of TMA production. The higher oxygen availability in this atmosphere on one hand may result useful in reducing the production of TMA, but on the other hand makes the carbon dioxide level to be lowered affecting its antimicrobial activity (Sivertsvik et al., 2002). The effect of modified atmosphere package on the fish shelf life and the most suitable mix composition for a specific application depends extensively on the fish species, fat content, initial microbiological contamination, background of the fish, treatment the fish undergoes after slaughtering, such us handling and storage condition, ratio of gas/product ($G/P$), and most importantly packaging method and temperature of storage (Bøknæs et al., 2000; Sivertsvik et al., 2002; Stammen et al., 1990). Thus, generalization cannot be drawn about shelf life extension and fish acceptability. Also, few information is reported in the literature about gutted and washed sea bass (Özogul, Gökbulut, Özyurt, & Özogul, 2005; Papadopoulos et al., 2003). Thus, the objective of this work was to explore how the different gas mixes influence the microbiological, chemical, physical and sensory indices of gutted farmed bass during chilling shelf life, with the main attributes of freshness being odour and appearance response.

## 2. Material and methods

### 2.1. Packaging and storage condition

Bass (*D. labrax*) were obtained from a local farm (ECOM (NA), Italy) during the period July–October 2004. The average bass weight was 190 g ranging from a minimum of 140 g to a maximum of 230 g. Fishes were slaughtered by immersion in ice-cold water (hypothermia), gutted and packed in an insulated polystyrene box with ice. Then, they were delivered to the laboratory within 2 h from the moment of the harvest. Once at the laboratory, fishes were washed, wiped, and modified atmosphere packed by using a packaging machine (Minipack Torre, TSM, 105, Cava dei Tirreni (SA), Italy). Fishes were packed with polystyrene tray laminated with a multilayer barrier film ($V = 1000$ cc; CoopBox, Bologna, Italy) and sealed with a film of PA/EVOH/PE ($P_{O_2} = 1.5$ cc/m$^2$ 24 h atm; $P_{H_2O} = 5.5$ g/m$^2$ 24 h atm; thickness = 55 μm). On the bottom of the tray was placed an absorbent layer to avoid excessive accumulation of exudates. The ratio between the volume of gas and weight of food product ($G/P$ ratio) was 2:1 (V/W). The gas mix compositions used to pack the product are listed in Table 1. All samples were stored in a refrigerator at constant temperature ($3 \pm 1$ °C) for 9 days. Analyses were performed immediately after packaging and after 2, 5, 7 and 9 days of storage.

### 2.2. Microbiological analysis

Standard enumeration methods were used to determine the microbial populations of the fish during chilled storage. Two samples of fish (25 g) were taken from each package, then diluted with 225 ml of peptone salt solution (0.85%NaCl–0.1%peptone) and homogenized with a Stomacher for 2 min. The sample was then serially diluted as needed for plating. The following media and incubation conditions were used: Plate Count Agar (PCA, Oxoid) for aerobic mesophilic bacteria (AMB) by pour plating, incubated at 30 ° for 72 h; violet red bile glucose agar (VRBGA, Oxoid) for *Enterobacteriaceae* (E) counts, incubated at 37 °C for 24 h. Microbial counts were performed in duplicate and expressed as cfu/g.

Due to the natural variability of the fresh samples, microbiological results were reported in terms of increment respect to the value at time zero (delta AMB; delta E).

Table 1
Headspace gas composition at the packaging time

| Atmosphere code | $O_2$% | $CO_2$% | $N_2$% |
|---|---|---|---|
| A | 0 | 70 | 30 |
| B | 20 | 70 | 10 |
| C | 30 | 60 | 10 |
| D | 40 | 60 | 0 |
| E | 30 | 50 | 20 |
| F | 21 | 0 | 79 |

### 2.3. Chemical analysis

*Gas analysis:* $O_2$ and $CO_2$ concentrations (% v/v) in the package head space were monitored by means of a portable PBI Dansensor A/S (Check Mate 9900 $O_2/CO_2$; Ringsted, Denmark) analyzer (accuracy $\pm$ 0.1%). 3 ml of gas were sampled from the package headspace with a needle.

*pH determination:* pH was measured by using samples obtained by mixing homogenized flesh fish with water (ratio 1:2) by using a pH meter Cyber Scan pH/Ion 510 (Eutech Instuments Pte Ltd. Ayer Rajah Crescent Singapore). The pH meter was equipped with a Schott electrode which was previously calibrated with buffer solutions (pH 4 and pH 7) at 20 °C. Four measurements were carried out on each samples.

### 2.4. Physical analysis

*Water loss:* The water loss of the samples was measured by gravimetric method. Raw bass was weighted before packaged and at each storage time, after it was removed from the packages. The difference of weight ($\Delta g$) was divided by the initial weight of the product (g) and expresses as $\Delta g/g\%$.

*Flesh moisture content:* The fish flesh moisture content was determined by gravimetric method. Flesh fish was dried in an oven at 100 °C and was accurately weighted at regular time interval until constant weight was reached. Three samples from each fish were measured. The moisture content was expressed as gram of water on gram of total weight ($g_{H_2O}/g_{tot}\%$).

*Colorimetric measurement:* The fish colour was measured with a colorimeter tristimolous (Minolta Chroma Meter, model CR-300) having a circular measurement area ($D = 8$ mm). The colorimeter was calibrated using a white standard plate ($L = 100$). The $L^*$, $a^*$ and $b^*$ values were measured on the abdomen, on the stomach and on the flesh of the gutted bass. Five readings were carried out at each position.

### 2.5. Sensory analysis

Sensory analysis was performed by a trained panel composed of 7 judges aging between 20 and 28 years. The panel training consisted of seven different sessions during which different samples were examined by the panel to select the most appropriate sensory attributes to describe bass freshness and to define their evaluation techniques (Table 2). During this stage, three different samples, in triplicate, were submitted to the panel to evaluate, also, the reproducibility of the judges answer and their capability in discriminating among samples. Next the panel was trained to assess each attribute by using a linear scale (Table 2). All the sample whose score was less than 50% compared to the score attributed at the fresh sample were classified as unacceptable. During the analysis, samples were presented in randomized order to minimize possible carry-over effects between different samples. During each session, judges were asked to analyze 6 different samples (2 different storage times, 2 different atmospheres, two fresh sample). All samples were analysed in triplicate. The test was carried out under white light. Fizz Network software (Biosystems, Counternon France) was used for data collection.

Table 2
List of sensory attributes for descriptive analysis of the gutted bass

| Attributes | | 0 | 5 | 10 |
|---|---|---|---|---|
| Odour | Odour at the pack opening | Spoilt odour | No odour | Fresh fish |
| Appearance | Eyes conformation | Concave | | Convex |
| | Flesh colour (belly cavity) | White | Pink[a] | Red |
| | Gill colour | White | Pink[a] | Red |
| Texture | Stiffness | Very slimy | Quite hard | Hard and elastic |

[a] The flesh and gill colour of the fresh fish was scored as 5.

### 2.6. Experimental design and data analysis

To study the effect of atmosphere and time on the bass quality attributes, a factorial design with two factors (time and atmosphere) was used. Six were the levels of the atmospheres (Table 1) and five were the levels of storage time (0, 2, 5, 7 and 9 days). Three replicates were performed for each experiment for a total of 90 samples. ANOVA analysis were performed on the data to evaluate the effect of atmosphere ($A$), time ($B$) and the interaction effect ($A \times B$) on the quality attributes. Duncan test were performed to find out the source of the significant differences within samples. Significance of differences was defined at $P \leqslant 0.05$.

All data were then used in principal component analysis (PCA) for classification of the samples on the basis of microbiological, chemical, physical and sensory data. The data matrix consists of 25 samples (rows) differing with respect to storage time and storage atmosphere. Composition column in the data matrix contained 19 quality attributes: pH, water loss, flesh moisture content, coordinates $L^*$, $a^*$, $b^*$ of the fish abdomen, stomach and flesh, odour, eyes conformation, stiffness, flesh colour, gill colour, AMB and *Enterobactriaceae*. Flesh colour and gill colour were scaled before PCA. All quality attributes were the average of the replicates.

## 3. Results and discussion

The ANOVA results showed a significant effect of the time and atmosphere on AMB and *Enterobacteriaceae* ($F = 20$, $P < 0.0001$; $F = 21$, $P < 0.0001$, respectively, for atmosphere and time).

The growth of AMB and *Enterobacteriaceae* was retarded when gutted bass was packed by using 20% of $O_2$ and 70% of $CO_2$ (B), and 30% of $O_2$ and 50% of $CO_2$

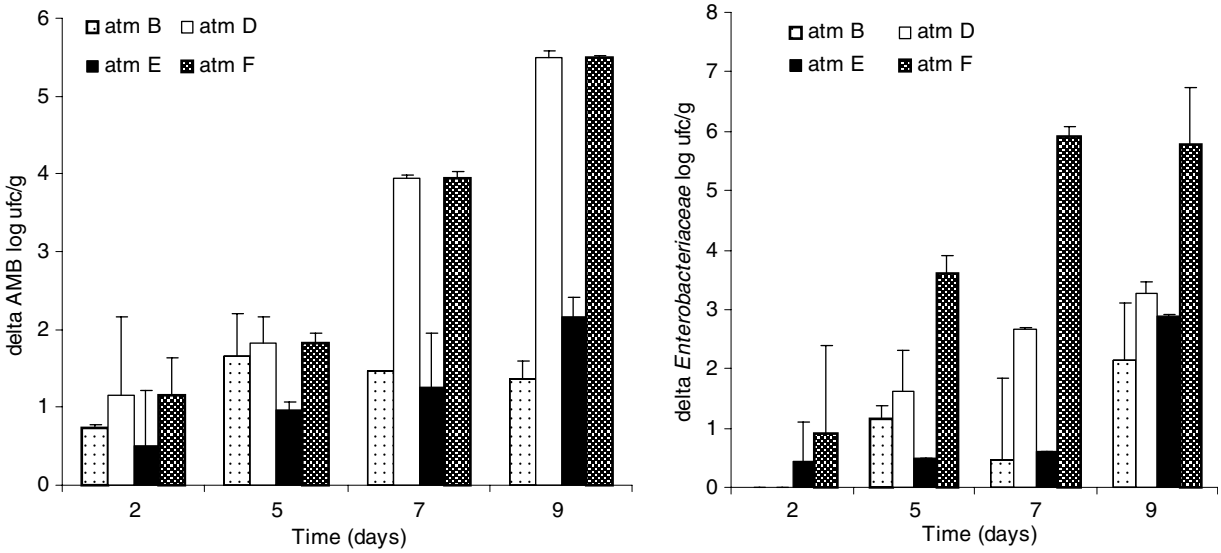


Fig. 1. Increment of AMB and *Enterobactericeae* of gutted bass during storage at 3 °C in MAP.

(E) (Fig. 1). After 9 days of storage at 3 °C AMB and *Enterobacteriaceae* reached, respectively, $2.5 \times 10^6$ cfu/g and $5.2 \times 10^4$ cfu/g for the sample B, and $2.2 \times 10^7$ cfu/g and $8.0 \times 10^5$ cfu/g for the sample E. On the other side, packing the gutted bass with air as initial gas composition (*F*), the contamination was more than $10^8$ cfu/g for the AMB and of about $10^7$ cfu/g for *Enterobacteriaceae* after 9 days of storage. By using 40%$O_2$–60%$CO_2$ (D), same results of sample F were obtained for AMB. This high growth rate of AMB may be due to the high level of oxygen in the package headspace. For all the above, it is possible to conclude that high level of carbon dioxide (>50%) have a protective effect on the product whether the level of oxygen is equal or less than 30%.

Headspace gas analysis could be an effective and easy index of microbial growth. In fact, in the case of the bass packed by using air as initial gas composition (*F*), oxygen decreases from 21% to 0.2% while carbon dioxide increases from 0% to 17% in 9 days (Table 3). The large variation of the gas composition of this samples is due to microbial growth, which on the contrary is inhibited by the high level of carbon dioxide in modified atmosphere package. The oxygen concentration of the gutted bass packed by using 20%$O_2$–70%$CO_2$ (B) and 30%$O_2$–50%$CO_2$ (E) showed, in agreement with microbiological results, a little oxygen decrease, respectively, after 7 and 9 days of storage. The only exception has been observed on the samples packed by using 40% of $O_2$ and 60% of $CO_2$ (D), whose oxygen concentration has remained almost constant during the storage period although microbiological results pointed out a growth of aerobic mesophilic and *Enterobacteriaceae*. In all the cases, carbon dioxide decreases probably as a consequence of the gas dissolution into tissue liquids (Ruiz-Capillas & Moral, 2001; Sivertsvik et al., 2004). Similar results were reported by Randell et al. (1997) which reported that oxygen concentration decreased and carbon

Table 3
Headspace gas composition of the gutted bass during storage at 3 °C

| ATM code | | Days of storage | | | | |
|---|---|---|---|---|---|---|
| | | 0 | 2 | 5 | 7 | 9 |
| A | $O_2$% | 1 ± 2 | 1.1 ± 0.3 | 1.4 ± 0.6 | 1.0 ± 0.3 | 1.5 ± 0.8 |
| | $CO_2$% | 70 ± 2 | 65 ± 1 | 63 ± 3 | 57 ± 3 | 60 ± 5 |
| B | $O_2$% | 20 ± 2 | 19 ± 1 | 20 ± 1 | 19 ± 1 | 14.5 ± 0.4 |
| | $CO_2$% | 70 ± 2 | 66 ± 3 | 70 ± 1 | 68.8 ± 0.8 | 57 ± 3 |
| C | $O_2$% | 30 ± 2 | 30.2 ± 0.7 | 29 ± 1 | 29.0 ± 0.5 | 27.9 ± 0.6 |
| | $CO_2$% | 60 ± 2 | 56 ± 4 | 54 ± 5 | 58 ± 4 | 59 ± 2 |
| D | $O_2$% | 40 ± 2 | 36.4 ± 0.5 | 37 ± 2 | 38 ± 2 | 37 ± 2 |
| | $CO_2$% | 60 ± 2 | 53 ± 2 | 52 ± 2 | 52.4 ± 0.8 | 49.9 ± 0.9 |
| E | $O_2$% | 30 ± 2 | 30 ± 1 | 30 ± 1 | 26 ± 2 | 13 ± 4 |
| | $CO_2$% | 50 ± 2 | 44 ± 5 | 40 ± 2 | 42 ± 3 | 40 ± 3 |
| F | $O_2$% | 20.8 ± 2 | 20.5 ± 0.7 | 4 ± 2 | 0.5 ± 0.5 | 0.2 ± 0.3 |
| | $CO_2$% | 0 ± 2 | 1.1 ± 0.7 | 13 ± 2 | 14.9 ± 0.8 | 17 ± 3 |

dioxide concentration increased during storage of MAP rainbow trout and Baltic herring as a result of microbial metabolism. They also reported that in the MAP package, carbon dioxide concentration decreased at the beginning of storage period, as $CO_2$ was absorbed by the fish, and then remained almost constant.

The positive effect due to high carbon dioxide concentration on the bacterial growth rate is paid of chemical and physical properties loss. Carbon dioxide dissolution into the tissue liquid leads to an acidification of the medium and the subsequent formation of carbonic acid. Hence pH will decrease (Colby & Flick, 1993; Sivertsvik et al., 2002), which in turn affects the water holding capacity and thus the texture properties of the product. Moreover, the belly flaps and the skin colour may be altered for whole fish stored in high $CO_2$ concentrations (Sivertsvik et al., 2002).

Fig. 2 shows the pH of the bass samples during storage as a function of the MAP composition. ANOVA results suggested that time, atmosphere and their interaction have a significant effect on bass pH change ($F = 124$, $P \ll 0.0001$; $F = 39$, $P \ll 0.0001$; $F = 25$, $P \ll 0.0001$, respectively). The fresh bass pH was equal to 6.6; once packed in modified atmosphere, the pH decreases to 6.5–6.3, depending on the storage atmosphere composition. Only the sample packed with air as initial gas composition ($F$) exhibited an increase of the pH which raised to a value of 6.84, as a consequence of the accumulation of basic substances in the bass muscles. Similar results were reported by Pastoriza et al. (1998) and Ruiz-Capillas and Moral (2001). From a practical point of view, it is worth to highlight that bass packed with 40% $O_2$ and 60% $CO_2$ (D) and 30% $O_2$ and 50% $CO_2$ (E) shows the lowest variation of pH during storage time. Those atmospheres were even the most protective one for the gutted bass water loss (Fig. 3). In particular, ANOVA results suggest that both time and atmosphere have a significant effect on the gutted bass water loss. However time has a more significant effect than atmosphere ($F_{5.60} = 10$, $P < 0.0001$; $F_{4.60} = 100$, $P < 0.0001$, respectively, for atmosphere and time effect) and interactions were found between the two effects ($F_{20.60} = 2$, $P = 0.008$). The effect of carbon dioxide on the water loss may depend on fish genera, $CO_2$ exposure and low temperature which may have a combined effect in function of specific MAP system (Stammen et al., 1990). In our experience water loss was reduced by lowering the level of carbon dioxide from 70% to 50%. From a practical point of view, even though MAP is not able to prevent drip formation, this inconvenient can be avoided by using a package with an absorbent bottom which helps reducing the drip accumulation on the package, overcoming the possible negative impact that the presence of drop may have on the consumer product acceptability. On the other hand, moisture content of the flesh fish did not change significantly during storage (Fig. 4) and the atmosphere composition had no effect on it ($P > 0.05$). Therefore one can conclude that water losses as small as 3–5% do not affect in a significant manner the juiciness of the flesh fish.

During storage time, colorimetric parameters related to the bass abdomen and stomach did not change (data not showed), while those related to the flesh did and their values at different storage time are reported in Table 4. ANOVA data suggested that only $a^*$ and $b^*$ are affected by the storage time, whereas $L^*$ is independent on time ($F_{4.21} = 0.6$, $P > 0.05$). The atmosphere composition has a significant effect on all the coordinate ($F_{5.30} = 2.5$, $P = 0.031$; $F_{5.11} = 3.5$, $P = 0.004$; $F_{5.97} = 14$, $P < 0.0001$, respectively, for $L^*$, $a^*$ and $b^*$). In particular, the parameter $a^*$ (redness) decreased during storage when bass

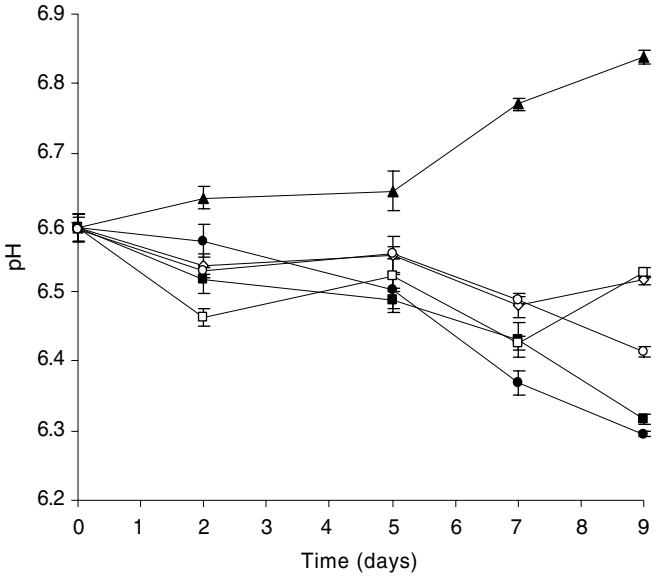


Fig. 2. pH of gutted bass during storage time in modified atmosphere package (atm A: □; atm B: ●; atm C: ○; atm D: ■; atm E: ◊; atm F: ▲).

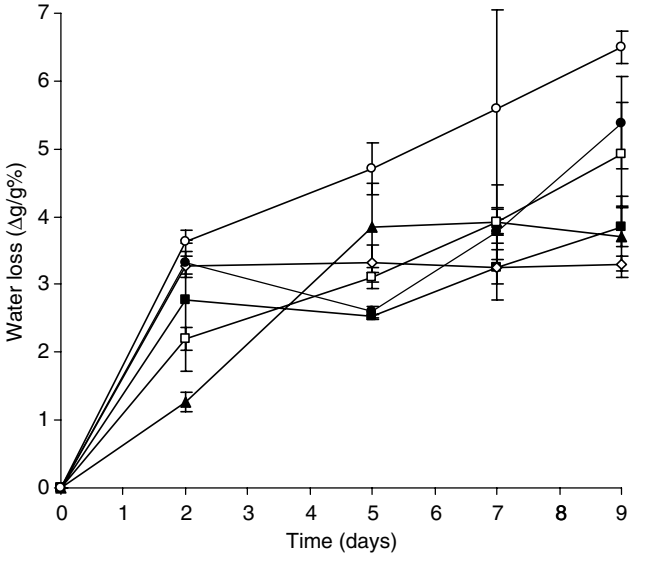


Fig. 3. Water loss changes of gutted bass during storage time in modified atmosphere package (atm A: □; atm B: ●; atm C: ○; atm D: ■; atm E: ◊; atm F: ▲).

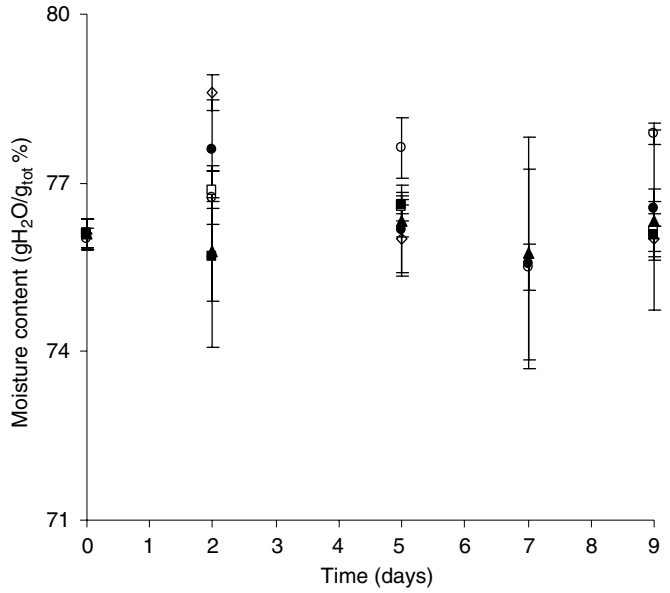


Fig. 4. Moisture content of gutted bass during storage time in modified atmosphere package (atm A: □; atm B: ●; atm C: ○; atm D: ■; atm E: ◊; atm F: ▲).

was packed by using 30% of $O_2$–60%$CO_2$ (C), 40%$O_2$–60%$CO_2$ (D), and 30%$O_2$–50%$CO_2$ (E), whereas it did not change when bass was packed by 70%$CO_2$–30%$N_2$ (A) and 20%$O_2$–70%$CO_2$ (B). The same was observed in the samples packed by using air as initial gas composition. Parameter $b^*$ (yellowness) increased during storage in the case of the atmosphere B, C and E, but decreased or remained constant in the other cases.

### 3.1. Sensory analysis

Sensory attribute scores given by the panel to the bass packed by using atmospheres having different composition are reported in Table 5. Time and atmosphere composition have a significant effect on all the attributes ($P < 0.0001$). During storage, the odour (evaluated as odour perceived at the opening of the package) changes with increasing the storage time passing from "fresh fish odour" (day 0–2) for all the samples to "spoilt odour" as a consequence of the extensive spoilage which had taken place. The effect of time is significant and can be appreciated since the second days of storage (Table 5). Only the odour of the sample packed by using air as initial gas composition (*F*) is perceived as off odour after 7 days of storage and after 9 days received a score as low as 0.91. Among the other samples, those made by using 20%$O_2$–70%$CO_2$ (B) and 30%$O_2$–50%$CO_2$ (E) received after 9 days of storage score higher than 5, i.e. the panel did not perceive any off odour at the package opening. With respect to appearance attributes, the eyes conformation is the only one which varies during storage, whereas flesh and gill colour change a little from pink. The eyes of the fresh fish is convex and became concave after 5 days. The bass packed by using the atmosphere with 30%$O_2$–50%$CO_2$ (E) is the one which received the best scores at each storage time. The colour of fresh bass was described as pink and to it corresponded a score equal to 5. Sample packed with air as initial gas composition (*F*) was the only one who preserved both the flesh colour and the gill colour during storage. Sensory results were in agreement with instrumental results which showed no variation of the chromatic coordinate of the samples F with varying the storage time. Instead, bass packed by using atmospheres rich in oxygen showed a whitening and a yellowing of both the flesh and the gill during storage time. By considering the appearance attribute, again atmospheres with 20%$O_2$–70%$CO_2$ (B) and with 30%$O_2$–50%$CO_2$ (E) resulted the most protective for the gutted bass.

The stiffness of the control samples changed during storage time and from hard and elastic it became very slimy.

Table 4
Colour evolution in gutted bass during storage at 3 °C for all the package conditions

| Hunter | Time/atm | A | B | C | D | E | F |
|---|---|---|---|---|---|---|---|
| $L^*$ | 0 | 65 | 65 | 65 | 65 | 65 | 65 |
| | 2 | 64 | 65 | 66 | 70 | 64 | 66 |
| | 5 | 66 | 61B | 70 | 68 | 60B | 67 |
| | 7 | 64 | 65 | 68 | 69 | 65 | 67 |
| | 9 | 66 | 68 | 69 | 63 | 65 | 61 |
| $a^*$ | 0 | 3.4 | 3.4 | 3.4 | 3.4 | 3.4 | 3.4 |
| | 2 | 2.9 | 2.7 | 2.2ab | 1.6b | 1.4b | 3.7 |
| | 5 | 2.5AB | 2.8AB | 1.8abBC | 1.2bBC | 4.0 | 3.6 |
| | 7 | 2.8 | 1.9 | 1.5ab | 0.7b | 1.4b | 2.9 |
| | 9 | 2.5AB | 1.2CD | 0.8bC | 0.2bCD | −1.0cD | 3.2 |
| $b^*$ | 0 | 1.1 | 1.1 | 1.1 | 1.1ab | 1.1 | 1.1 |
| | 2 | −1.0bcC | 1.0BC | 4b | −1.2cC | 3.4AB | 1.1C |
| | 5 | −1.3cB | 4.3B | 3.6b | 1.3abAB | 4.0 | 1.3AB |
| | 7 | −1.0bcD | 3.8AB | 5.4b | 0.6bcCD | 3.8AB | 1.5CD |
| | 9 | 1.0ab | 2.6 | 3.1b | 3b | 3.4 | 0.8 |

Statistics in small letters compare the storage time in each atmosphere and parameter.
Statistics in capital letters compare the different atmosphere for each storage time and parameter.

Table 5
Sensory attribute scores of gutted bass in different modified atmosphere packaging during storage at 3 °C

| Parameters | Time | Atmospheres | | | | | |
|---|---|---|---|---|---|---|---|
| | | A | B | C | D | E | F |
| Odour | 0 | 9.45 | 9.45 | 9.45 | 9.45 | 9.45 | 9.45 |
| | 2 | 8.16bAB | 8.1bAB | 7.9bB | 8.0bB | 8.5b | 8.0bB |
| | 5 | 7.40bAB | 6.9cAB | 6.0dC | 6.8cAB | 7.6b | 6.0cC |
| | 7 | 4.25cB | 6.3c | 6.3c | 5.2dAB | 6.4c | 2.4dC |
| | 9 | 4.30cB | 6.4c | 6.3c | 4.0cB | 6.1c | 0.9eC |
| Eyes conformation | 0 | 9.16 | 9.16 | 9.16 | 9.16 | 9.16 | 9.16 |
| | 2 | 4.6cC | 7.3bAB | 8.1bAB | 6.9bC | 7.6bAB | 8.4 |
| | 5 | 6.2bAB | 5.7cB | 3.4cC | 5.8cB | 7.0b | 4.5bC |
| | 7 | 2.7dB | 3.6eAB | 3.2cB | 3.4dAB | 4.4c | 2.8cB |
| | 9 | 2.1dB | 4.3d | 2.5dB | 2.5eB | 4.1c | 2.8cB |
| Flesh colour | 0 | 4.94 | 4.94 | 4.94 | 4.94 | 4.94 | 4.94 |
| | 2 | 4.9 | 4.6abAB | 3.6bB | 4.0bAB | 4.5abAB | 4.9b |
| | 5 | 4.3cAB | 3.7cdBC | 2.7cC | 3.4cBC | 4.4abAB | 4.9b |
| | 7 | 4.1cd | 3.9bcBC | 3.6bBC | 5.3cC | 3.8bBC | 4.7c |
| | 9 | 4.6b | 3.4dBC | 2.4cBC | 1.4dC | 2.5cBC | 4.8c |
| Gill colour | 0 | 5 | 5 | 5 | 5 | 5 | 5 |
| | 2 | 4.2bB | 4.3bB | 4.7AB | 4.0bB | 4.0bB | 4.7 |
| | 5 | 5 | 3.9bcB | 2.9cC | 3.9bB | 2.9cC | 4.9 |
| | 7 | 4.5ab | 3.1dB | 3.5bAB | 3.6bAB | 1.9dC | 3.9bB |
| | 9 | 4.1bAB | 3.5cdBC | 1.4dD | 2.2cCD | 2.0dCD | 5 |
| Stiffness | 0 | 9.15 | 9.15 | 9.15 | 9.2 | 9.15 | 9.15 |
| | 2 | 6.4bCD | 8.1bAB | 5.9bD | 7.6bABC | 7.3bBCD | 8.5 |
| | 5 | 6.4bBC | 6.7cABC | 4.3cD | 6.2cC | 7,3bAB | 7.6b |
| | 7 | 3.0cC | 6.7c | 4.2cBC | 5.1dB | 6.7b | 3.1cC |
| | 9 | 2.7cCD | 4.9dB | 3.5cC | 2.7eCD | 6.7b | 2.3dD |

Statistics in small letters compare the storage time in each atmosphere and parameter.
Statistics in capital letters compare the different atmosphere for each storage time and parameter.

The best score relative to stiffness was received by the bass packed by using 20%$O_2$–70%$CO_2$ (B) and 30%$O_2$–50%$CO_2$ (E), whose flesh stiffness remained almost constant during storage time.

The effect of time and atmosphere on the sensory attributes is well depicted by the two principal components plot obtained by PCA analysis (Fig. 5). PC1 is mainly defined by stiffness, eyes conformation and odour parameters,

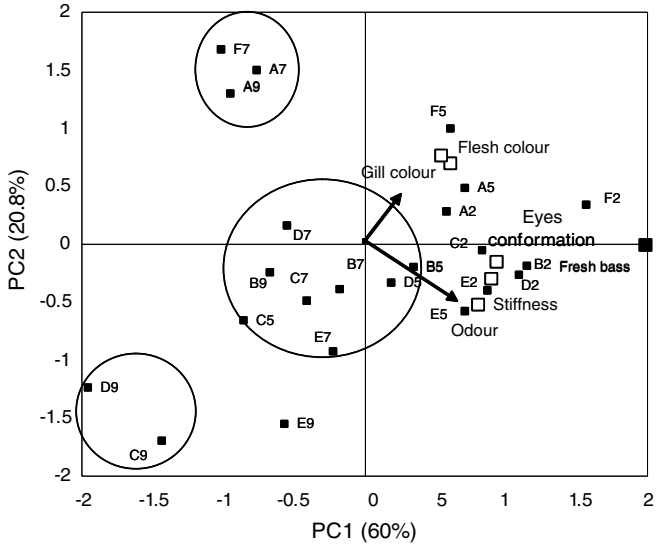


Fig. 5. Bi-plot of PCA scores and loadings characterizing gutted bass samples and sensory attributes, respectively. Letters indicate bass packed with different atmosphere (tab.1), number indicate the storage time.

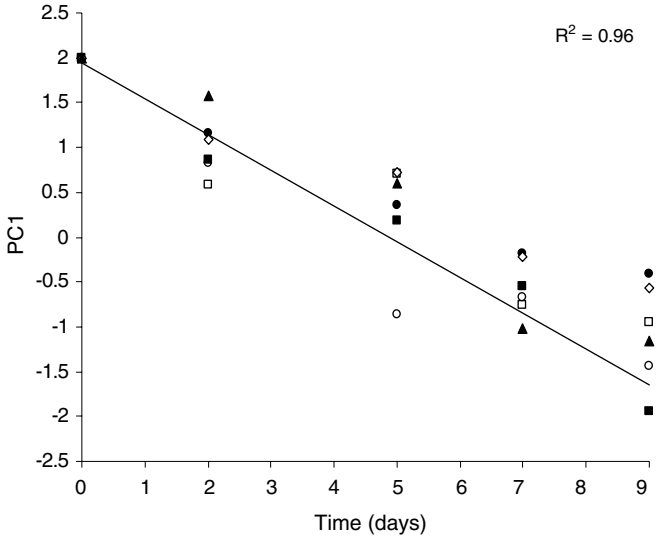


Fig. 6. Evolution of factor 1 (sensorial parameters) on storage time according to the different modified atmosphere packaging (atm A: □; atm B: ●; atm C: ○; atm D: ■; atm E: ◊; atm F: ▲).

and PC2 by appearance parameters (flesh and gill colour). Together they explain 88% of variance. Factor 1 is basically linear related to the length of storage period, as suggested by the uniform distribution of the subjects along this axis. Fig. 6 helps understanding better the relationship between factor 1 and storage time. By contrast, the distribution of the samples along the factor 2 axis shows quite clearly that the samples position is related to their mode of storage (Fig. 5). In particular, fresh sample which received the highest scores for the attribute: odour, eyes conformation and firmness, is located on the right side in the region corresponding to the positive values of the PC1, while, with increasing the storage time, all the samples move from the right side to the left one of the plot, toward negative PC1 value which are negatively related to freshness attributes. Moreover, with increasing the storage time, the position of samples packed by using 20%$O_2$–70%$CO_2$ (B), 30%$O_2$–60%$CO_2$ (C), 40%$O_2$–60%$CO_2$ (D), and 30%$O_2$–50%$CO_2$ (E) changes mainly because of gill and flesh colour, while for samples packed by using 70%$CO_2$–30%$N_2$ (A) and 21%$O_2$–79%$N_2$ (F) because of stiffness, eyes conformation and odour attributes The freshness of bass samples as evaluated by means of sensory analysis and given by the factor 1 decreases as time increases for all the samples (Fig. 6). By submitting microbiological, sensory and instrumental parameters together to Factor analysis, the results shown in Figs. 7a and b were obtained.

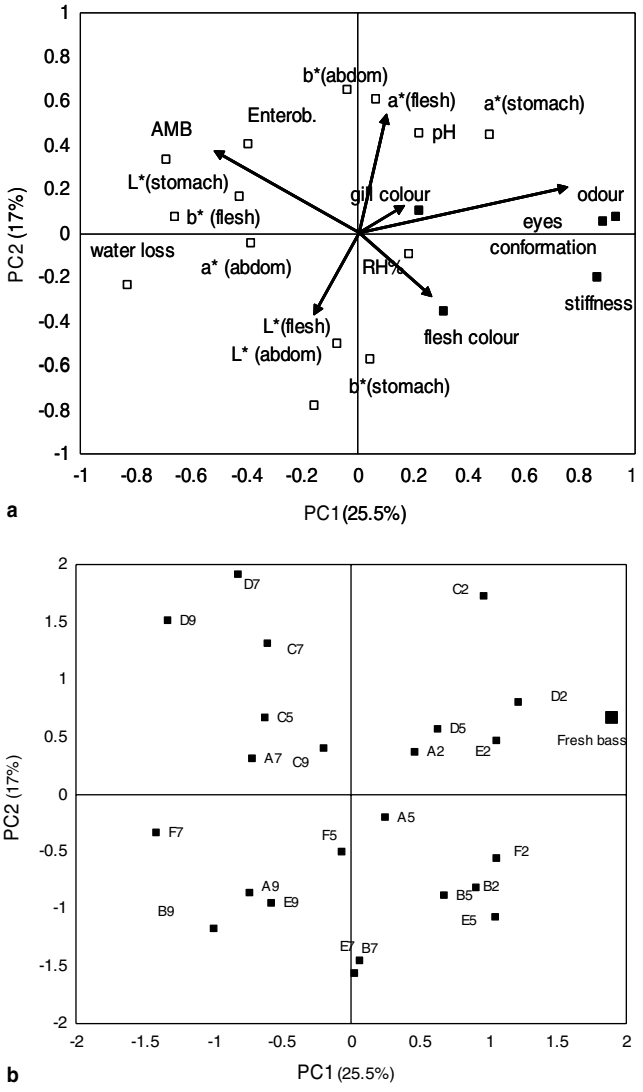


Fig. 7. Loading (a) and score (b) plot of the first two Factors of instrumental (□) and sensorial (■) parameters of gutted bass in modified atmosphere package. Letters indicate bass packed with different atmosphere (Table 1), number indicate the storage time.

Principal components 1 and 2 explained 25.5% and 17% of the total variation of the data, respectively.

Flesh and gill colour evaluated by the trained judges are correlated positively with instrumental $L^*$ and $a^*$ of the flesh. Water loss is negatively related to stiffness, pH and moisture content (RH%). Microbiological results (AMB, *Enterobactericeae*) are high related with odour evaluated by sensory analysis: higher is the contamination of the product, lower is the odour sensory score. Fig. 7b is in agreement with the conclusion derived by describing Fig. 5, i.e. the samples distribute them self along the factor 1 axis depending on the storage time and along the factor 2 depending on the atmosphere composition.

## 4. Conclusions

Modified atmosphere packaging with gas mixture rich in carbon dioxide is an effective method to preserve microbiological quality of reared and gutted bass during storage at low temperature (3 °C). Although very high level of carbon dioxide favours water losses, this can be reduced by almost 50% by reducing the carbon dioxide concentration from 70% to 50%. In addition, by using packs with absorbent bottom, it is possible to avoid the presence of visible moisture in the package. High oxygen level (>30%) promoted yellowing of the bass flesh and gill more than oxygen free atmosphere or air. By considering a shelf life of 7–9 days, the best results in terms of microbiological, chemical, physical and sensory attributes are guaranteed by gas mixtures composed of 30% of oxygen and 50% of carbon dioxide. By means of Principal Component Analysis was possible to clarify the contribute of the atmosphere composition and time on the quality changes of bass: factor 1 describes the quality evolution with varying the time; factor 2 describes the quality evolution in function of the atmosphere composition.

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