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Food Chemistry 74 (2001) 317–325

Food Chemistry

www.elsevier.com/locate/foodchem

Chilled bulk storage of gutted hake (*Merluccius merluccius* L.) in CO_2 and O_2 enriched controlled atmospheres

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Received 29 February 2000; received in revised form 12 February 2001; accepted 12 February 2001

Abstract

This work compares hake gutted whole and placed in boxes in ice under controlled atmospheres (CA) with four different gas mixtures: M1 $CO_2/O_2/N_2$(%) (60/15/25), M3 (40/40/20), M4 (60/40/0) and M5 (40/60/0) and hake in air for 33 days of storage. The storage chamber temperature was $0 \pm 1°C$. Biochemical analyses [pH, total volatile basic nitrogen (TVB-N), trimethylamine nitrogen (TMA-N) and thiobarbituric acid (TBA index) and sensory analyses (inspection and tasting)] were carried out to study the quality of the hake. The hake, kept under controlled atmospheres, did not exceed the limits for acceptability of 35 mg/100 g and 12 mg/100 g of TVB-N and TMA-N, respectively, throughout the storage period, and the TBA values remained lower than the recommended limits of 14 mg of malonaldehyde/100 g. Chemical results accorded with the tasters who did not detect any off-odour in the samples, and the M4C lots stored under 60/40/0 ($CO_2/O_2/N_2$%) was rated the best. However, this result did not accord with the inspection or pH by which all the lots were rejected at day 25 of storage. Despite this, the atmosphere system for preservation of chilled hake could be used safely and effectively on fishing boats to extend the shelf life of this species, and was better than conventional storage in ice.

Keywords: Gutted hake; Bulk storage; Controlled atmosphere; Shelf-life; Spoilage index

1. Introduction

Hake (*Merluccius merluccius* L.) consumption in Spain has increased in recent years. This species, although primarily marketed fresh, is also marketed frozen, but to a slightly less extent. Consumers of this fish demand intrinsically high-quality hake with excellent sensory characteristics. Traditionally, storage of fresh hake has been in ice and a refrigerated atmosphere. However, this system only keeps the fish fit for human consumption for a relatively short period of time, depending on the species, type of capture and season (Stammen, Gerdes, & Caporaso, 1990). Hake, and fish in general, deteriorate very quickly after capture due to enzymatic autolyses *and* post-mortem changes in pH which potentiate microbial growth (Ward & Baj, 1988). Hence the need for a storage system, such as MAP, which preserves the quality and maintains the hygienic, sanitary and organoleptic characteristics of this fresh fish. The fish species, and its initial quality, will govern the success or failure of the storage system used and will therefore partly determine the shelf life of the fish in controlled or modified atmospheres.

There are different ways of applying modified or protective atmospheres: modified atmosphere packaging (MAP), controlled atmosphere packaging (CAP) or controlled atmosphere storage (CAS), and equilibrium modified atmosphere (EMA) packaging; vacuum packing, hypobaric storage and hyperbaric storage (Phillips, 1996; Wilhelm, 1982). The principal gases used for modified atmospheres are CO_2, O_2 and N_2. These gases have been used in different combinations and proportions, depending on the product and process requirements, and whether it is packaged or in bulk (Church & Parsons, 1995; Farber, 1991; Phillips, 1996). Atmospheres with low O_2 and high concentrations of CO_2 in relation to air have been used to delay the growth of microorganisms. Studies by Dainty (1971) reported that CO_2 concentrations at 5–10% inhibit the growth of many bacteria, especially aerobes, which grow on fish stored at refrigerated temperatures and which are primarily responsible for fish spoilage. However, there are many studies with atmospheres using CO_2 concentrations, as high as 100% (Lannelongue, Hanna, Finne,

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0308-8146/01/$ - see front matter
PII: S0308-8146(01)00159-5

Nickelson, & Vandirzant, 1982; Parkin & Brown, 1983; Silva, Harkness, & White, 1993). More recent studies have revealed that high CO_2 concentrations are not so beneficial, whereas an increase in the O_2 concentration in the mixture might be more appropriate (Lannelongue et al., 1982, López-Gálvez, De La Hoz, & Ordóñez, 1995). In many studies (Fey & Regenstein, 1982; Genigeorgis, 1985; Gray, Hoover, & Muir, 1983), O_2 atmospheres have been used to prevent the growth of *Clostridium botulinum*, which grows on fresh fish if it is at high temperatures. In some previous works, atmospheres with high O_2 concentrations were assayed, with very positive biochemical results (Ruiz-Capillas, 1997).

The storage of fish in modified atmospheres has traditionally been used for preservation in packages (Banks, Nickelson, & Finne, 1980; Handumrongkul & Silva, 1994; Lannelongue et al, 1982; Phillips, 1996; Strenstrom, 1985). This procedure consists of replacing the the air in the container around the fish with a gas or a gas mixture and the modifications which occur inside the container are left unaltered during the storage period. On the other hand, for controlled atmospheres, which have been used less (Fujii, Hirayama, Okuzumi, Yasuda, Nishino, & Yokoyama, 1990; Kimura, Kuroda, Murakami, & Fujii, 1996; Woyewoda, Bligh, & Shaw, 1984) as well as placing the air inside the container, the mixture is maintained and regulated accordingly. In other words, the controlled atmospheres regulate the gas used and the humidity very closely and a monitoring system keeps these parameters within very strict margins (Oberlender, Hanna, Miget, Vanderzant, & Finne, 1983). The advantages of using a controlled atmosphere are to ensure a more stable composition of the gases throughout the storage and avoid the accumulation of off-odours during storage in modified sealed atmospheres. In controlled atmospheres, the off-odours are eliminated by the gas flow through the container (Oberlender et al., 1983). The application of controlled atmospheres could also be of great interest for fish preservation on deep-sea fishing boats, such as those which go to Morocco and Grand Sol and fish in tides for 15 to 20 days, and are very important for the Spanish economy. Fish, and particularly hake landed by these boats, is kept in ice using the traditional system. The quality of the fish captured during the first few days of fishing may then not be fit for human consumption when the boat reaches port. Thus, the use of onboard controlled atmospheres would be of great interest for prolonging the shelf life of the landed fish. Furthermore, with this onboard storage system, the demand for fish in the market could possibly be regulated for 1 or 2 weeks according to demand.

Bearing in mind these considerations, the effect of controlled gas atmosphere, using different CO_2 and O_2 concentrations, on the shelf life of gutted and whole hake was studied. Biochemical and sensory tests were carried out to study the effectiveness of these mixtures.

2. Materials and methods

The species used for this work was hake (*Merluccius merluccius* L.) because of its great commercial importance in Spain. Hake was used the day it was caught by dragnet from the Galician shelf. Hake were selected on the boat, gutted and placed in boxes with ice. When the boat reached port, the boxes of hake were sent to the Instituto del Frío (IF; Madrid) in an isothermal lorry with plenty of ice. The different lots were immediately prepared after the fish arrived at the IF. The 170 kg consignment of hake was divided into five lots. One lot was kept in the boxes with ice as in conventional storage. The other four were each placed inside different hermetic stainless steel containers ($78\times48\times56$ cm) and these were closed. Then, through the orifices, in the sides of the containers, the four gas mixtures: Mixture 1 (60/15/25, $CO_2/O_2/N_2$), mixture 3 (40/40/20, $CO_2/O_2/N_2$), mixture 4 (60/40, CO_2/O_2) and mixture 5 (40/60, CO_2/O_2) were injected into each, from a pressurised bottle, during 30 min to a pressure of 1 bar until the concentration of gases inside was close to the injected mixture with a variation of 10%. Once the mixtures had stabilised, measurements and flushings were done every day to keep the atmosphere controlled. The containers were placed in a forced ventilation chamber at a programmed temperature of $0\pm1^{\circ}C$ together with the control lot. The boxes of hake were kept in the containers and the fish were analysed every week until they deteriorated.

2.1. Measurement of temperature

Temperatures were measured every 6 h with copper thermocouples connected to a Yokogawa Hokushin Electric meter, model 3087, Tokyo (Japan) and into the container atmosphere, and the hake in the container, and likewise for the control hake and in the chilled room.

2.2. Measurement of gases

Controls of the CO_2 and O_2 gases were performed every 12–24 h for the first few days it was necessary to carry out an analysis every 12 h, since the CO_2 concentrations decreased quickly and it was necessary to inject fresh mixture. Once the atmosphere in the containers was stable, the analysis of gases was performed every 24 h with an ABISS PACK 12 apparatus (France) which determined the amount of CO_2 and O_2 simultaneously.

2.3. Biochemical analyses

2.3.1. Biochemical analyses

At least five individuals were taken from each lot, cut into slices from which skin and bone were removed, and then all the muscle was macerated to obtain homogeneous

representative samples. These analyses were normally done in triplicate.

2.3.2. Determination of pH

The muscle was homogenised in water in a ratio 1:10 (v/w) and measured using a Radiometer model PHM 93 pH-meter at ambient temperature.

2.3.3. Determination of the 2-thiobarbituric acid (TBA) index

This was determined by the method of Vyncke (1970) as modified by Lemon (1975).

2.3.4. Determination of total volatile basic nitrogen (TVBN)

This was determined by the method Antonocopoulos and Vyncken (1989). The distillation was performed in a Tecator model 1002.

2.3.5. Determination of trimethylamine nitrogen (TMAN)

This was determined by the method of the AOAC (1995).

2.4. Sensory analysis

Two forms of sensory analysis were used, namely inspection and taste panel. Inspection was performed by three trained persons on at least five samples of hake from each lot. Each sample was classified using an analysis of anatomical and histological parameters: general appearance, skin, gill colour and odour and eyes. Each characteristic was scored on a scale of 0 to 10, with 10 representing a fresh fish and 0 a putrified fish, and lots scoring less than 4 were rejected. An average score for all observed parameters was made on each sample. Taste panels were carried out in a tasting room built to the specifications in standard proposal UNE 33119 (1976). Measurements were performed by a panel of seven semi-trained tasters selected from I.F. personnel. The samples had first been cut into slices, placed on a tray, covered and placed in a microwave for 6 min, after which they were presented to the panel. The panel assessed the following parameters: colour, odour, flavour, watery, firmness and general acceptability. Points were awarded on a scale of 0 to 10, and lots scoring less than 4 were rejected. The methodology followed for analysis of inspection and test panel was according to Ruiz-Capillas (1997).

2.5. Statistical analysis of results

The differences between the different lots for the different parameters were determined by multifactorial analysis of variance (random block model) to determine whether there were differences between the treatments, and which of the treatments was best suited to hake. Differences between pairs were resolved by least significant difference (LSD) test to establish confidence intervals. The level of significance was set at 95%.

3. Results and discussion

The average temperature of the forced ventilation chambers (where the control and containers were kept in the experiments) was $0.34\pm0.3^{\circ}C$. The average temperatures inside the containers where the hake was under the effect of the atmosphere, and the hake inside were $-0.32\pm0.2^{\circ}C$ and $-0.63\pm0.1^{\circ}C$, respectively. The temperature of the control iced hake in boxes in the same chamber as the containers was $0.25\pm0.1^{\circ}C$. These temperatures are within the optimum range for the preservation of chilled fish (Fell, 1991). This range of temperatures was used to increase the effect of the atmospheres. Exhaustive control of the temperatures improves the biochemical and sensory results for the lots kept in atmospheres and the control lot.

The CO_2 and O_2 measurements inside the containers, for each of the mixtures studied during storage time, are shown in Figs. 1–4. For all the lots, the CO_2 levels were slightly lower than the concentration injected with the mixture. Moreover, this decrease in CO_2 levels might as well be due to CO_2 absorption and dissolution in the tissue liquids, as other authors have highlighted (Lannelongue et al., 1982). The oxygen levels, however, were very similar to the injected mixtures, except for the mixture M5C which had 60% oxygen. Thus, it would appear that the greater the levels of a gas in the mixture, the more difficult it is to keep these levels constant when the atmospheres are applied. This was observed in previous studies (Ruiz-Capillas, 1997). The use of controlled atmospheres ensures that the gas composition is more stable throughout the storage, thereby guaranteeing effective treatment (Oberlender et al., 1983).

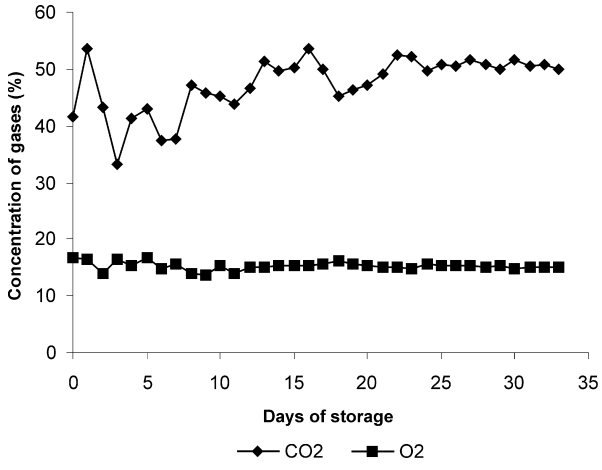

Fig. 1. Evolution of concentration of CO_2 and O_2 in the container of lot M1C storage in controlled atmosphere with the mixture M1 (60/15/25, $CO_2/O_2/N_2$%).

Fig. 5 shows the pH results for the different hake sample lots kept under controlled atmospheres. The initial pH in hake was 6.88. Villenure, Simard, and Picard (1986) found initial pH values in gutted cod of 6.82, very similar to the values found in this study on hake. The control lots showed a slight increase during all the storage; however, a significant difference ($P \leqslant 0.05$) was observed only on day 33 when it reached a pH of 7.5 (Fig. 5). The significant differences ($P \leqslant 0.05$) between the lots in atmospheres and the control lot appeared at day 5 of storage, because the pH in the lots kept in controlled atmosphere decreases until day 19 or 25 of storage and then increases, but only slightly (Fig. 5). This decrease in pH could be due to acidification of the medium from CO_2 dissolution in the tissue liquids and the formation of carbonic acid (Banks et al., 1980). This has also been reported by other researchers, such as Banks et al. (1980) and Woyegoda et al. (1984). It accorded with the decrease observed in the evolution of CO_2 in the containers (Figs. 1–4). The increase in pH in the lots in atmospheres from day 25 of storage was more pronounced in lot M4C, where the pH reached 7.33 after 33 days of storage, and there were no significant differences ($P \leqslant 0.05$) between lots and control. The other lots, kept under atmospheres, were not significantly different ($P \leqslant 0.05$) from one another and pH values were < 7. The significant increase in pH is due to the accumulation of basic substances in the hake muscle (Banks et al., 1980; Hebard, Flick, & Martin, 1982). However, the results for the pH in this

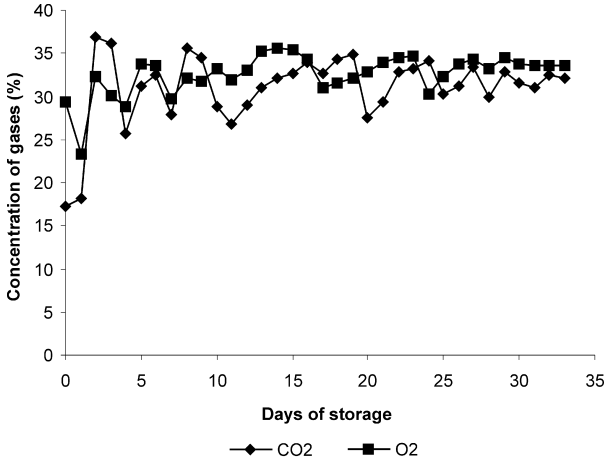

Fig. 2. Evolution of concentration of CO_2 and O_2 in the container of lot M3C storage in controlled atmosphere with the mixture M3 (40/40/20, $CO_2/O_2/N_2$%).

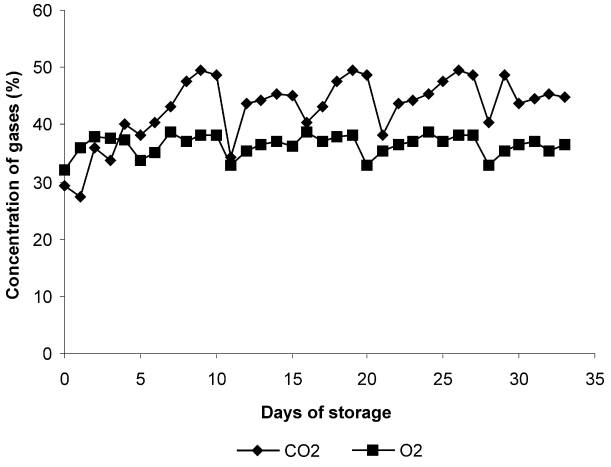

Fig. 3. Evolution of concentration of CO_2 and O_2 in the container of lot M4C storage in controlled atmosphere with the mixture M4 (60/40/0, $CO_2/O_2/N_2$%).

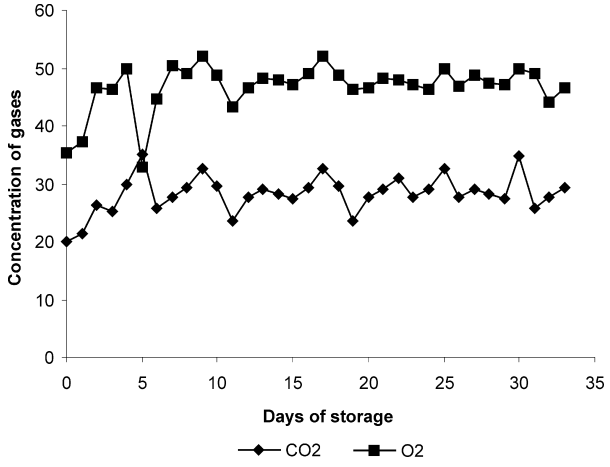

Fig. 4. Evolution of concentration of CO_2 and O_2 in the container of lot M5C storage in controlled atmosphere with the mixture M5 (40/60/0, $CO_2/O_2/N_2$%).

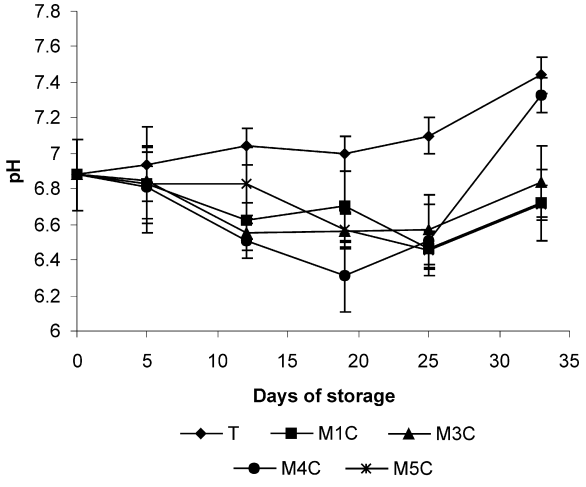

Fig. 5. pH of hake stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C). T, control lot, stored in air throughout the experiment.

experiment did not accord with the results of TVBN and TMA-N, which presented lowest levels in the lot MC4 (Figs. 7 and 8, respectively).

The lipids of fresh fish, kept in ice or in refrigeration storage, rarely have any tendency towards oxidative rancidity (Liston, Stansby, & Olcott, 1963). Nevertheless, when this fish is stored for long periods of time in controlled atmospheres with high O_2 levels, rancidity could be a problem. However, in this study no logical correlation was observed between O_2 levels in the assayed atmospheres and TBA levels during the period studied (Fig. 6). Lot M5C, kept in the atmosphere with the greatest O_2 concentrations (60%), had TBA levels of 0.97 mg of malonaldehyde/100 g, after 25 days of storage,

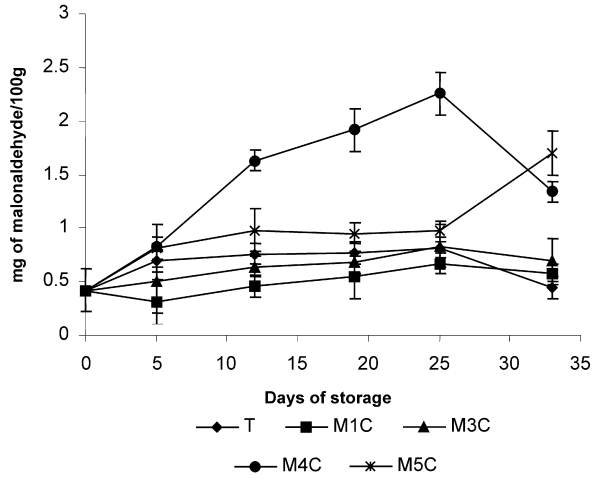

Fig. 6. 2-thiobarbituric acid (TBA) (mg of malonaldehyde/100 g) of hake stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C). T, control lot, stored in air throughout the experiment.

lower than lot M4C, which had 40% O_2. Perhaps this is due to synergistic action in the CO_2 and O_2 concentrations to facilitate polysaturated fatty acid autooxidation (Fig. 6). In all the lots, TBA values stayed quite constant and there were no significant differences ($P \leqslant 0.05$) until day 25 of storage except for the lot M4C (Fig. 6). In this lot, TBA values fluctuated during storage but remained within the recommended limits of 14 mg of malonaldehyde/100 g of muscle (Connell, 1975). In the other lots, there was no significant ($P \leqslant 0.05$) increase for the storage period studied, indicating that lipid oxidation is not of major importance in this study. Fey and Regenstein (1982), in a study on red hake and salmon stored in modified atmospheres, also suggested that the changes in TBA are not important. The tendency of TBA to increase in lot M4C throughout the storage could be the reason for the inspection result on day 12 of storage when the hake odour and gill colour were identified as slightly rancid, which coincided with a sharp increase in TBA levels in this lot. However, for this same 12-day period, the tasters did not detect any off-odour or taste in these samples. Other authors have also observed similar behaviour in modified atmospheres (Brown, Albright, Watts, Heyer, Spruce, & Price, 1980)

The TVB-N and TMA-N values increased significantly ($P \leqslant 0.05$) during storage (Figs. 7 and 8); this increase was much slower in the lots in controlled atmospheres than in the control lot. Villemure et al. (1986) also observed that TVB-N levels in gutted cod, stored in CO_2 atmospheres, increased more slowly than lots in air. The control lot had the highest TVB-N values throughout the storage period, exceeding the limits for acceptability of 35 mg/100 g of muscle indicated in EEC Directive 95/149 after 25 days of storage, whereas the lots in the different atmospheres did not

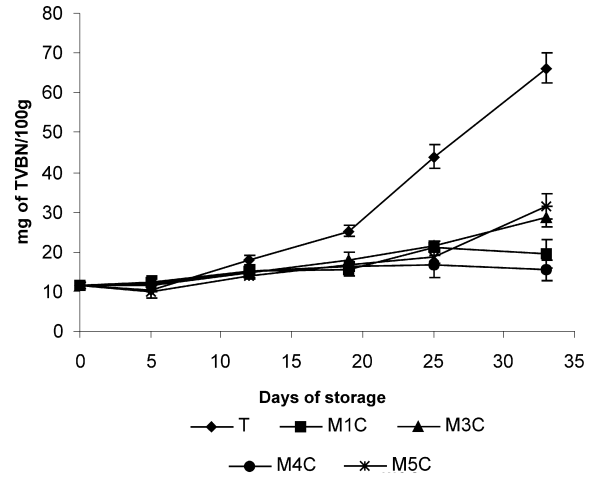

Fig. 7. Total volatile basic nitrogen (TVBN) (mg/100 g) of hake stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C). T, control lot, stored in air throughout the experiment.

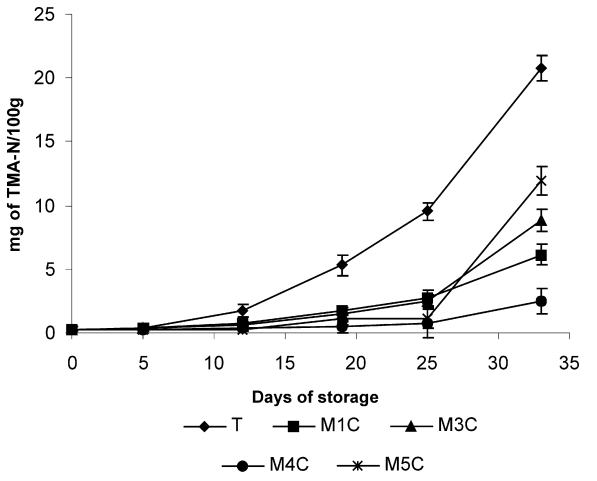

Fig. 8. Trimethylamine nitrogen (TMAN) (mg/100 g) of hake stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C). T, control lot, stored in air throughout the experiment.

exceed this limit throughout the storage period (Fig. 7). Similar behaviour was observed for the TMA-N values, where the control lot had higher values than the treated lots and the control lot exceeded the limits of 12 mg/100 g of muscle indicated in EEC Directive 91/493 from day 25 of storage (Fig. 8). According to some authors (Banks et al., 1980; Lampila, 1991; Lannelongue et al., 1982; Mockhele, Johnson, Barrett, & Ogrydziak, 1983; Parkin, Wells, & Brown, 1981), this could be due to two factors: on the one hand, the inhibiting effect of CO_2 on microbial growth, and on the other, the change in flora when fish is stored in enriched CO_2 atmospheres.

The TVB-N and TMA-N values in the control lot were significantly different ($P \leqslant 0.05$) from the other lots in atmospheres after day 12 of storage (Figs. 7 and 8, respectively). The lots in controlled atmospheres had constant TVB-N and TMA-N levels until day 25 of storage and significant differences ($P \leqslant 0.05$) between them appeared at day 33 of storage. The lots kept in atmospheres with greater CO_2 concentrations (M1C and M4C) are those which had the lowest TVB-N and TMA-N values, especially lot M4C that was also kept with high CO_2 and O_2 concentrations (60/40%, respectively). This lot after 33 days of storage had TVB-N and TMA-N values of only 15.4 mg/100 g and 2.49 mg/100 g, respectively. This may be due to the fact that the controlled atmospheres delayed the onset of spoilage and the subsequent TVB-N and TMA-N substances, unlike the control lot. However, this result did not accord with that observed in the inspection (Table 1), where all the lots, on general appearance, were rejected at day 25 of storage, and there were no significant differences ($P \leqslant 0.05$) between the control lot and the treated ones in this period. For the parameters: gill colour, odour and eye, the worst-rated lots were the binary atmospheres M4C and M5C, which were rejected, together with the control, after 12 days of storage while the lots M1C and M3C were rejected day at 25 (Table 1). The emission of unpleasant odours in hake are due to the accumulation of volatile compounds, a consequence of bacterial growth, mostly TVB-N and TMA-N (Huss, 1995; Lyndsay, Josephaon, & Olafsdottis, 1986). However in the lots kept in controlled atmospheres, the production of these substances was very low (Figs. 6 and 7).

Table 1
Sensory evaluation (Inspection) for hake stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C)[a]

Parameters	Lots	Days of storage					
		0	5	12	19	25	33
General Appearance	T	10±0.0	9.3±0.4	4.6±0.9	5.0±0.6	0.4±0.3	0.0±0.0
	M1C		7.7±1.0	6.5±0.6	7.8±0.9	3.0±0.9	1.6±0.8
	M3C		8.3±0.9	8.9±0.7	7.8±0.8	1.9±0.8	1.1±0.8
	M4C		9.7±0.8	6.5±0.7	5.2±1.1	2.6±0.7	1.8±0.7
	M5C		8.4±0.6	4.6±0.8	4.1±0.8	3.6±0.9	3.0±0.9
Skin	T	9.9±0.1	8.5±0.8	3.9±0.7	5.5±0.7	5.0±0.8	0.0±0.0
	M1C		7.6±0.7	7.0±1.1	8.3±0.9	3.3±0.9	0.8±0.5
	M3C		9.1±0.6	7.8±0.9	4.0±0.6	2.0±0.8	0.8±0.6
	M4C		7.7±0.8	7.3±0.8	4.0±0.7	4.6±0.9	5.2±0.8
	M5C		7.7±0.9	4.5±0.8	4.0±0.6	6.8±0.8	3.0±0.9
Gills colour	T	9.6±0.3	8.7±0.8	6.6±0.7	2.8±0.8	2.9±1.1	0.0±0.0
	MIC		7.7±1.1	9.0±0.6	2.6±0.7	3.2±0.8	1.6±0.8
	M3C		9.5±0.4	4.5±0.9	2.6±0.9	3.2±0.8	0.5±0.3
	M4C		8.2±0.9	1.0±0.5	0.2±0.1	4.4±0.9	2.2±0.8
	M5C		8.2±0.7	4.2±0.8	1.3±0.5	4.4±0.7	2.2±0.7
Odour	T	9.6±0.4	6.3±0.6	1.5±0.8	0.0±0.0	0.9±0.7	0.0±0.0
	M1C		8.1±0.8	7.7±1.1	7.8±0.9	0.0±0.0	0.5±0.3
	M3C		9.6±0.3	7.0±0.9	7.1±0.8	0.0±0.0	1.1±0.8
	M4C		5.0±0.7	2.4±0.7	3.1±0.8	3.7±0.8	1.0±0.9
	M5C		6.3±0.6	3.9±0.8	3.1±0.7	3.7±0.7	1.0±0.7
Eyes	T	9.5±0.4	4.4±0.8	4.4±0.9	0.3±0.2	0.0±0.0	0.0±0.0
	MIC		4.5±0.7	5.8±0.7	2.8±0.8	1.4±0.6	0.0±0.0
	M3C		4.5±0.8	5.8±0.8	2.8±0.7	1.4±0.8	0.0±0.0
	M4C		6.0±0.7	2.1±0.7	0.6±0.4	1.5±0.7	0.0±0.0
	M5C		6.0±0.5	3.0±0.6	0.6±0.4	0.6±0.5	0.0±0.0

[a] T, control lot, stored in air throughout the experiment. Reject from 4 points (10, excellent and 0 very bad).

Table 2
Sensory evaluation (taste panel) stored during 33 days in controlled atmosphere (C) with four different gases M1 (60/15/25, $CO_2/O_2/N_2$%) (M1C), M3 (40/40/20) (M3C), M4 (60/40/0) (M4C) and M5 (40/60/0) (M5C)[a]

Parameters	Lots	Days of storage				
		0	5	12	19	25
Colour	T	10±0.0	7.8±0.9	7.0±0.8	5.9±0.8	–
	M1C		8.2±0.8	7.1±0.9	5.6±1.0	4.2±0.8
	M3C		8.3±0.8	6.6±0.8	4.0±0.7	5.1±0.7
	M4C		6.9±0.7	5.1±0.6	6.7±0.6	6.4±0.9
	M5C		6.3±0.6	5.4±0.7	5.5±0.6	6.3±0.9
Odour	T	9.7±0.2	7.0±1.1	6.0±0.8	4.4±0.9	–
	MIC		6.8±0.9	5.8±0.8	5.2±0.8	2.3±0.8
	M3C		6.2±0.8	6.1±0.9	4.7±0.7	3.0±0.4
	M4C		6.4±0.6	6.2±0.7	5.9±0.6	5.3±0.8
	M5C		6.4±0.7	5.7±0.6	5.2±0.6	5.3±0.7
Flavour	T	9.8±0.1	7.1±0.9	7.3±0.8	5.1±0.8	–
	MIC		7.3±0.9	6.4±0.7	5.3±0.7	2.6±0.6
	M3C		7.7±0.6	7.0±0.9	3.9±0.6	3.1±0.8
	M4C		7.1±0.7	5.7±0.8	6.2±0.9	5.6±0.7
	M5C		7.5±0.8	6.1±0.7	4.5±0.8	5.4±0.9
Wateriness	T	10±0.0	4.8±1.1	4.9±1.0	4.9±0.8	–
	MIC		7.6±0.8	5.1±0.9	6.6±0.9	3.1±0.8
	M3C		4.5±0.8	6.4±0.9	4.5±0.9	3.6±0.7
	M4C		6.4±0.6	5.3±0.7	5.9±0.7	3.0±0.7
	M5C		7.1±0.7	6.2±0.6	5.5±0.6	2.8±0.6
Firmness	T	9.8±0.2	6.2±0.9	5.8±0.8	4.6±0.8	–
	MIC		7.4±1.1	6.3±0.7	5.4±0.7	3.6±0.9
	M3C		7.7±0.8	5.1±0.9	6.2±0.6	3.1±0.9
	M4C		5.5±0.9	5.3±0.8	5.5±0.8	6.6±0.8
	M5C		5.4±0.8	4.1±0.7	5.1±0.9	5.8±0.7
General acceptability	T	9.8±0.1	6.3±0.8	5.3±0.4	4.8±0.9	–
	MIC		7.2±0.7	5.2±0.8	4.7±0.8	2.6±1.1
	M3C		6.8±1.0	6.6±0.9	4.4±0.9	2.1±0.9
	M4C		6.9±1.0	5.6±0.7	5.8±1.1	4.0±0.8
	M5C		7.3±0.9	5.4±0.7	4.7±0.5	3.3±0.7

[a] T, control lot, stored in air throughout the experiment. Reject from 4 points (10, excellent and 0 very bad).

Moreover, these atmospheres might possibly be used for storing hake fillets, where the negative market aspects of eye and gill colour would not be an issue.

On the other hand, the tasting analysis presented a very good relation with the TBA, TMA-N and TVBN but not with the inspection analysis and pH. The results of this test panel showed that lot M4C was rated best, especially on the colour of the flesh parameter, as happened in the case of TMA-N and TVBN. The wateriness parameter received the lowest score and all were rejected on this basis at the end of the storage (Table 2). This latter phenomenon has also been observed by other authors and they have explained that it is due to excessive exudation in fish in modified atmospheres resulting from a loss in its capacity to retain water (Cann, 1984; Pastoriza, Sampedro, Herrera, & Cabo, 1996; Tiffnety & Mills, 1982). However, in this instance the lowest score for wateriness was in the control lot. After 25 days of storage, lots M5C and M4C received the highest score for all the parameters studied and were not rejected on the basis of colour, odour, flavour or firmness, unlike lots M1C and M3C

4. Conclusion

All the lots were rejected much sooner by the sensory inspection analysis than would be expected from the biochemical and tasting analyses. The lots kept under controlled atmosphere were not rejected during the 33

days of storage; however, from the inspection analyses, all the lots were rejected at 25 days of storage. However, the use of controlled atmospheres throughout the storage of hake, with high O_2 concentration (40%), enhanced the biochemical quality of the hake as long as they were also accompanied by high CO_2 concentrations (60%). Moreover, the use of the atmosphere rich in O_2 did not cause any rancidity problem.

Acknowledgements

The experiment reported here is a part of the project "Maintaining quality of fresh fish aboard ship and on land by modified atmosphere. Development of equipment, containers and procedures" contract no. AIR1 CT92-0273 EU and ALI93-1004-CE (C.I.C.Y T.). We would like to thank Air Liquid (SL, Spain) for the supply of gases employed in this work.

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