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Application of modified atmosphere packaging on aquacultured fish and fish products: A review

Achilleas D. Bouletis^a, Ioannis S. Arvanitoyannis^a, and Christos Hadjichristodoulou^b

^aSchool of Agricultural Sciences, Department of Agriculture, Ichthyology and Aquatic Environment, University of Thessaly, Volos, Hellas, Greece;

^bFaculty of Medicine, Laboratory of Hygiene and Epidemiology, University of Thessaly, Hellas, Greece

ABSTRACT

The aquaculture industry has undergone a rapid and continuous growth during the last decade due to increased demands in seafood consumption and reduced pelagic fish production. The aquaculture sector can provide products with a consistent flow and quality standards to cover the market's needs. The modified atmosphere packaging (MAP) technique has evolved over the last two decades and many reports indicate its beneficial effect on many quality parameters during fish and shellfish preservation. The use of MAP can clearly offer an advantage to the safer distribution of quality aquacultured fishery products. This article summarizes most of the experimental data of packaging techniques applied (MAP, VP, various pretreatments and packaging materials) on aquacultured fish and fish products to provide a clear view of the potential for a future commercial use.

Abbreviations: HP: High Pressure; MAP: Modified Atmosphere Packaging; TVC: Total Viable Count; TMA-N: Trimethylamine-Nitrogen; TVB-N: Total Volatile Base- Nitrogen; TBA: Thiobarbituric acid; LAB: Lactic Acid Bacteria; PCB: Polychlorinated biphenols; PMAP: Passive MAP; G/P: Gas/Product; ASC: Acid-soluble collagen; ISC: Insoluble collagen; PP: Sodium Pyrophosphate; APC: Aerobic Plate Count; FFA: Free Fatty Acids; TPC: Total Plate Count; MDA: Malondialdehyde; AMAP: Active Modified Atmosphere Packaging; TBARS: Thiobarbituric Reactive Substances; VP: Vacuum Packaging; PP: Pyrophosphate; UV: Under Vacuum; TSP: Trisodium Phosphate; PSC: Pepsin-soluble collagen; TSP: Trisodium Phosphate; STPP: Sodium Tripolyphosphate

Keywords

Aquacultured; fish; modified atmosphere; vacuum; packaging; microorganisms; sensory; physico-chemical properties

Introduction

Aquaculture is defined as the farming of aquatic organisms including finfish and shellfish, either individually or at industrial scale with the use of several interferences (feeding, confinement, and breeding) to maximize production. There is historic evidence for aquaculture in Egypt and China from 2500 and 100 BC, respectively (Sapkota et al., 2008). Aquaculture, including all types of culture of marine life, shows high potential and is one of the most developing food producing sectors worldwide (Defoirdt et al., 2011). The aquatic products coming from aquaculture techniques reached 148 million tons per year in 2010, of which 128 million tons were destined as food while preliminary data state that in 2011 the production increased to 154 million tons (FAO, 2012).

One of the main factors potentially affecting the quality and the safety of the products is the continuous monitoring and administration of the breeding environment (Paterson et al., 1997). The aquatic environment allows the growth of pathogenic bacteria at greater extent than the terrestrial environment and can reach high populations in their host animals (Defoirdt et al., 2011). In a study conducted by Wong et al. (1999) *V. parahaemolyticus* was detected in 315 out of 686 (45.9%) seafood samples imported from Asian countries. Another issue that

concerns the aquaculture industry is the consumer protection from health hazards such as the generation of pathogens that are resistant to antibiotics and can be transmitted through improper cooking to humans, or the cumulative effect of antibiotics, organochlorine compounds and metals (Grigorakis and Rigos, 2011). This phenomenon is further enhanced by rising temperatures due to global warming. In sea bream samples preserved at different temperatures for three months there was a significant temperature pending on accumulation of cadmium (Cd), copper (Cu), mercury (Hg), zinc (Zn), lead (Pb) and iron (Fe) in liver and Mn, Fe and Zn in fish muscle (Guinot et al., 2012). Manmade contaminants were detected in salmon [pesticides, polybrominated biphenol ethers (PBBEs), and polychlorinated biphenols (PCBs)], in sea bass (PCBs) and dioxins in catfish. The health risk due to consumption of contaminants may overshadow the benefits on the cardiovascular system that consumers have from certain farmed fish (Cole et al., 2009).

The quality of the products can also be affected by the harvesting method used, triggering biochemical processes that can lead to deterioration of flesh characteristics (Paterson et al., 1997). The initial state of freshness can be modified through the *post mortem* deterioration processes. Handling, storage conditions and the procedure that the fish undergoes after slaughtering can be referred as key factors along with the fish

species and history that can determine the occurrence and the severity of those phenomena (Cakli et al., 2006). Furthermore, the impact of killing method on quality of the product is another issue to be taken into consideration. Although most researchers focus on the stress induced during killing, handling prior to killing (crowding) may also have a drastic impact on fish (Poli et al., 2005; Bagni et al., 2007; Grigorakis, 2010). The conditions of breeding such as exercise can result in changes of muscle composition and consequently in higher flesh quality (Palstra and Planas, 2011).

The issue of commercialization of aquacultured transgenic animals due to enhanced production efficiency also poses many questions about the fish welfare and the customers' safety. Fish with elevated growth hormone increased the rates of protein synthesis and lipid mobilization thereby affecting factors like metabolic rates and body composition (Hallerman et al., 2007).

Modified atmosphere packaging technology is mainly based on substitution of the surrounding atmosphere of the product with a gas mixture that will provide the desirable effects like microbial growth delay, retarding the TMA and TVBN formation or sustenance of sensorial characteristics. Therefore, MAP use on fishery products can substantially lengthen product's shelf life, reduce potential economic losses, cut down distribution costs due to shipments reduction because of the substantial shelf life extension and lead to the supply of high quality products (Sivertsvik et al., 2002 cited in Arvanitoyannis and Stratakis, 2012). Moreover, the product can be easily evaluated by the potential customer due to transparent packaging materials. With these attributes MAP fulfills the most important functions of food packaging like protection of the product against deteriorative effects, efficient containment, use of product as a marketing tool and finally, an easy to use and convenient product (Yam et al., 2005).

Due to the complexity of the aquaculture procedure and in order to reduce the microbial and physiological deterioration of the samples many technologies were applied. MAP was one of the techniques widely tested for the preservation of many aquatic products. The bactericidal effect of CO₂ and the reduced physiological changes caused by atmosphere modification can play an important role in preserving quality and extending the shelf life of aquacultured fish and fish products. The aim of this review is to provide a thorough literature survey referring to MAP applied on aquacultured fish and fish products.

Sea bass

An active atmosphere modification (10% O₂/80% CO₂/10% N₂) was applied to evaluate the effect on chemical parameters of sea bass stored at 4°C. Ca²⁺, Mg²⁺, and Mg²⁺-Ca¹⁺ ATPase activity of natural actomyosin remained unaffected that is 0.31, 0.36, and 0.4 μmol Pi mg/protein.minutes, respectively, on the 21st day on samples stored under MAP, while there was a slight increase in Mg-EGTA ATPase (0.1 μmol Pi mg/protein.minutes). The sulfhydryl content dropped at a lower rate compared to that of control sample (5.5 mol10⁻⁵. g/protein) (Masniyom et al., 2004).

Masniyom et al.(2005a) investigated collagen changes in sea bass fillets preserved under MA conditions (10% O₂/80% CO₂/10% N₂) after pyrophosphate (PP) pretreatment at 4°C for 21 days. MAP prevented the occurrence of any changes made

in acid soluble and pepsin soluble collagen with the pretreatment having no effect on their contents; 0.17 and 0.1 g/100 g, respectively, for both untreated and treated samples under MAP by the end of storage. The maintenance of collagen on MAP samples at the end of storage is a clear indicator that MAP samples with or without PP pretreatment maintained firmness close to the initial levels (a 14.2% decrease in breaking strength on samples stored under MAP with or without PP pretreatment compared to 54.7% in air).

Three different pretreatments [trisodium phosphate (TSP), sodium pyrophosphate (PP) and sodium tripolyphosphate (STPP)] combined with MAP (10% O₂/80% CO₂/10% N₂) were used for sea bass preservation at 4°C for 21 days. All pretreatments gave similar results in reducing microorganism numbers of TPC and psychrophiles (6 and 5.8–6 log CFU/g, respectively) whereas there was no reduction in LAB (3.8 log CFU/g). The increase in TMA and TVB was controlled on pretreated MAP samples with all treatments leading to similar results (0.19 and 0.045 and 0.29 and 0.17 mg/g muscle for control and MAP samples, respectively) (Masniyom et al., 2005b).

Addition of thyme essential oil to sea bass samples stored under two different atmosphere modifications (10% O₂/40% CO₂/50% N₂ for MAP 1 and 10% O₂/60% CO₂/30% N₂ for MAP 2) at 4 ± 0.5°C for 21 days. Samples under MAP 2 with thyme oil displayed prolonged shelf life by 12 days in terms of APC (Control, air + thyme oil, MAP 1 and MAP 2 samples reached the acceptability limit of 7 log CFU/g for APC after 7, 9, 10, and 12 days of storage, respectively). The limit for TVB-N was reached after 6, 8, 9, 13, and 17 days of storage for control, air+thyme oil, MAP 1, MAP 2 and MAP 2 + thyme oil, respectively (Kostaki et al., 2009).

Provincial et al. (2010) investigated the shelf life extension of sea bass samples stored under MAP (40% CO₂/60% N₂ for MAP 1, 50% CO₂/50% N₂ for MAP 2 and 60% CO₂/40% N₂ for MAP 3) at 4°C for 21 days. TBARS values were similar in MAP samples whereas differed significantly with control samples (0.11–0.21 mg MDA/kg at the end of storage for MAP samples). APC and psychrotrophic growth were both limited by the application of MAP (6.05, 6.5, and 7.28 log CFU/g for APC and 7.06, 7.62, and 8.39 log CFU/g for psychrotrophs for MAP 3, 2, and 1, respectively). Shelf life was extended up to 11 days for MAP 1 and 14 days for samples stored under MAP 2 and 3.

The antimicrobial effect of irradiation (3 kGy) for quality improvement of sea bass samples or stored under MAP (20% O₂/40% CO₂/45% N₂ for MAP 1 and 5% O₂/60% CO₂/35% N₂ for MAP 2) at 2 ± 1°C for 10 days was assessed by Reale et al.(2008). Irradiation had a detrimental effect on microorganism growth and along with samples stored at MAP 2 had the best microbial quality; 1 and 5.6; 5.4 and 4.8; 5.9 and 7.6; 6.5 and 7.5; 5.3 and 5.5; 4.1 and 5.7; and 1 and 6.9 log CFU/g for LAB, *Pseudomonas spp.*, mesophiles, psychrotrophs, *Enterobacteriaceae*, total coliforms and *B. thermosphacta*, respectively.

The use of both passive and active MAP [5% O₂/70% CO₂/25% N₂ for MAP 1 with a perforated polystyrene (PS) tray and a Cryovac lid and 15% O₂/63% CO₂/22% N₂ for MAP 2 with a rigid tray] for the preservation of sea bass at 3°C was compared with vacuum packaging on a research conducted by Mercogliano et al. (2009). The shelf life was extended up to 12 days in terms of sensory

characteristics for samples stored under MAP 2 and PMAP. Biogenic amines (putrescine, cadaverine, spermidine, and spermine) were reported 13, 5.3, and 3 mg/kg for PMAP and 9.5, 5.5, 15, and 0 for MAP 2 on the 22nd day from catch, respectively

Gutted sea bass was stored under various O₂ and CO₂ concentrations (70% CO₂/30% N₂ for MAP 1, 20% O₂/70% CO₂/10% N₂ for MAP 2, 30% O₂/60% CO₂/10% N₂ for MAP 3, 40% O₂/60% CO₂ for MAP 4 and 30% O₂/50% CO₂ for MAP 5) at 3°C for nine days. Microorganism numbers were limited more effectively under MAP 5; that is 2.2×10^7 and 8×10^5 CFU/g for APC and *Enterobacteriaceae*, respectively. MAP 5 was shown to be the best in preserving sensorial and chemical quality of sea bass (Torrieri et al., 2006).

An experiment on storage of whole gutted or filleted sea bass samples stored on ice (whole samples) or under MAP (40% CO₂/60% N₂ for the sea bass fillets) at $2 \pm 1^\circ\text{C}$ was carried out by Poli et al. (2006). MAP and iced samples displayed lower microorganism counts compared to control (5.83 and 5.6 for TVC, 4.58 and 5.39 for *Pseudomonas spp.* and 5.74 and 4.37 log CFU/g for H₂S-producing bacteria, respectively), while there were different results in malonaldehyde content (0.33, 0.14, and 0.13 mg/kg for MAP, iced and control samples, respectively). The iced samples displayed the longest shelf life period reaching 10 days after slaughtering compared to eight and seven days for MAP and control samples, respectively.

Seven different atmosphere compositions (60% CO₂/40% N₂ for MAP 1, 10% O₂/60% CO₂/30% N₂ for MAP 2, 20% O₂/60% CO₂/20% N₂ for MAP 3, 80% CO₂/20% N₂ for MAP 4, 10% O₂/80% CO₂/10% N₂ for MAP 5, 20% O₂/80% CO₂ for MAP 6 and 100% CO₂ for MAP 7) were tested by Masniyom et al. (2002) for preservation of sea bass fillets stored at 4°C for 21 days. Microorganisms growth changed inversely with the CO₂ concentration, with MAP 7 having the lowest numbers (5.8 and 6 log CFU/g for APC and LAB on the 21st day). Samples stored under CO₂-rich atmospheres were acceptable in terms of TMA and TVB content by the end of storage (0.042 and 0.19 mg/g for MAP 7). Sea bass samples stored under MAP with 80–100% CO₂ displayed a shelf life that could be extended to more than 20 days.

The application of brine super-chilling (-1.5°C) prior to MAP (30% O₂/40% CO₂/30% N₂) storage was tested on *Labrax Japonicus* fillets by Liu et al. (2010). All the spoilage factors monitored during the experiment (TVC, H₂S-producing bacteria, TVB-N, TBA and myofibril fragmentation index) improved on the pretreated samples compared to MAP samples (6.4 and 6.9 log CFU/g, 5.9 and 6.45 log CFU/g, 20 and 25 mg/100 g, 1.45 and 1.85 mg MDA/kg and 135 and 160 for pretreated MAP samples and untreated ones, respectively). The combination of MAP and super-chilling extended shelf life by four days compared to MAP stored samples.

An atmosphere high in CO₂ (10% O₂/80% CO₂/10% N₂) and a pretreatment with pyrophosphate were used to control the growth of the inoculated *L. monocytogenes* and *E. coli* O:157 strains (10^3 and 10^5 log CFU/g) on sea bass fillets stored at 4°C for 21 days. There was a 1.4 to 1.5 log CFU/g reduction of *Listeria* populations at both inoculation levels on the pretreated MAP samples. *E. coli* populations on the samples treated displayed a substantial reduction at both inoculation levels reaching 1.6 and 3 log CFU/g on the 21st day of storage (Masniyom et al., 2006).

NaCl used for salting smoked sea bass fillets MA (70% CO₂/30% N₂) or vacuum packaged was replaced partially (100% NaCl or 50% NaCl/50% KCl was used) with KCl prior to storage at 4°C for 42 days. The combination of salts did not have any beneficial effect on microbial load, TVB-N, TMA and TBA values and sensory scores. Biogenic amine formation was delayed because of salt synergistic action (3.1, 22.1, and 6 mg/kg for histamine, putrescine and cadaverine values of samples stored under MAP) (Fuentes et al., 2011) (Table 1).

Mussels

Mussels were vacuum-packed (VP) or stored under MAP (20% O₂/60% CO₂/20% N₂ for MAP 1 and 40% CO₂/60% N₂ for MAP 2) for 14 days at $3 \pm 0.5^\circ\text{C}$. Sensory attributes were better preserved under MAP 1 (10–11 days) while MAP 2 also led to a shelf life prolongation of the products (seven to eight days). TMA and TVB-N of samples stored under MAP 1 were the lowest (11 and 36 mg N/100 g, respectively) but TBA was significantly higher than the other treatments (1.38 mg MDA/kg). The TVC of samples under MAP 1 was 6.7 log CFU/g, without exceeding the acceptability limit of 7 log CFU/g (Goulas, 2008).

The maintenance of quality and longevity of live mussels stored under MA environments (5% O₂/20% CO₂/75% N₂ for MAP 1 20% O₂/50% CO₂/30% N₂ for MAP 2, 50% O₂/50% N₂ for MAP 3, 75% O₂/25% N₂ for MAP 4 and 100% O₂ with a partial vacuum leading to an atmosphere of 70–80% O₂) at 2–3°C for six days was investigated by Pastoriza et al. (2004). The use of MAP 4 resulted in a shelf life extension of 48–72 hours with improved chemical (10.42 mgTVB-N/100 g for TVB at day six) and microorganism parameters (4.27 log CFU/g).

The microbial, biochemical and sensory changes of mussels stored under MAP (50% CO₂/50% N₂ for MAP 1, 80% CO₂/20% N₂ for MAP 2, 30% O₂/40% CO₂/30% N₂ for MAP 3) and VP at 4°C for 15 days were monitored by Goulas et al. (2005). An 1, 0.7, 0.7, and 1 log CFU/g reduction in TVC, *Pseudomonas*, H₂S-producing and LA bacteria was recorded on samples under MAP 2 at the 15th day. TVB and TMA for MAP 2 samples remained lower than the acceptability limits (25.22 and 7.87 mg N/100 g, respectively, at the end of storage) The longest shelf life extension obtained was six to seven days under MAP 2.

Wild mussels were packaged under MAP (50% CO₂/50% N₂ for MAP 1, 80% CO₂/20% N₂ for MAP 2, 65% CO₂/35% N₂ for MAP 3) and VP and kept under refrigeration ($2 \pm 1^\circ\text{C}$) for 12 days. MAP 2 gave the best microbiological results (6.8, 6.78, and 5.42 log CFU/g for TPC, psychrophilic bacteria and LAB, respectively). Chemical parameters remained acceptable until the eighth day of storage (30.6 mg/100 g, 3.3 mg malondialdehyde/kg and 4.2 mg/100 g for TVB-N, TBA and TMA-N, respectively) (Caglak et al., 2008).

Different sizes of live mussels (33 and 44 units/kg) were stored under various MAs (75% O₂ for MAP 1 and 85% O₂ for MAP 2) at $2 \pm 1^\circ\text{C}$ for days. The smaller mussels of higher ammonium and volatile fatty acid content were negatively affected as in-package O₂ increased. The survival rate of 20% exceeded for small mussels on day 13 for both modified atmospheres whereas on larger mussels samples were rejected before day 13 for MAP 2 (Bernardez and Pastoriza, 2011).



Table 1. A synopsis of the results of various MAP storage conditions on shelf life and microbial, chemical and sensory evaluation of sea bass samples.

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)— shelf life extension.	Refs
Sea bass.	80% CO ₂ /10% O ₂ /10% N ₂ . The fish/gas ratio was 1:3.	Film with OTR: 46.6 cm ³ /m ² /day.	4°C/21 days.	Ca ²⁺ , Mg ²⁺ , Ca ²⁺ , ATPase activities of natural atomyosin (NAM) did not present any noteworthy changes on MAP samples until the end of the experiment. Total sulphydryl content showed a smaller decrease on samples stored under MAP compared to control whereas surface hydrophobicity had an evident increase.				Masniyom et al., 2004.
Sea bass (<i>Lates calcarifer</i>) muscle.	80% CO ₂ /10% O ₂ /10% N ₂ . The fish/gas ratio was 1:3.	Film with OTR: 46.6 cm ³ /m ² /day. Treatment with Sodium Pyrophosphate.	4°C/21 days	Acid-soluble collagen (0.17 g/100 g) (ASC) and pepsin-soluble collagen (0.1 g/100 g) (PSC) did not significantly change on MAP samples with or without Sodium Pyrophosphate pretreatment. A slight decrease on insoluble collagen (ISC) was observed on MAP samples.		MAP treated samples with or without PP treatment preserved their firmness attributes better compared to air samples (54.7 and 14.2% firmness decrease for control and MAP samples, respectively, on day 21).		Masniyom et al., 2005a.
Sea bass slices	80% CO ₂ /10% O ₂ /10% N ₂ . The fish/gas ratio was 1:3.	Film with OTR: 46.6 cm ³ /m ² /day. Pretreatment with three different phosphate compounds including trisodium phosphate (TSP), sodium pyrophosphate(PP) and sodium tripolyphosphate (STPP).	4°C/21 days	Reduced changes in sulphydryl content and surface hydrophobicity detected on samples pretreated with PP proved its effectiveness in retarding protein denaturation. TVB content on samples under MAP was limited under 0.20 mg/g. Treatment with phosphates increased water uptake and consequently reduced exudate loss.	The synergistic effect of MAP and PP was evident in reducing mesophile microbial load (5.4 log CFU/gr). Psychrophilic bacterial (5.8 and 3.7 log CFU/g for psychrophiles and LAB, respectively) counts and LAB were lower in samples under MAP and PP than those of other samples.	MAP and PP pretreatment proved to be effective in reducing exudate losses and restraining water uptake ability while treated samples had the highest odor and flavor scores compared to both control and MAP samples without PP treatment.		Masniyom et al., 2005b.
Organically aquacultured sea bass (<i>Dicentrarchus labrax</i>) filets.	1. 40% CO ₂ /10% O ₂ /50% N ₂ .	Addition of thyme (T) oil (0.2% v/w), used as a natural preservative.	4 ± 0.5°C/21 days.	A shelf life of 6, 8, 9, 13, and 17 days for control, control and T, MAP 1, M AP 2 and MAP 2 and T,	Pseudomonads, LAB and H ₂ S producing bacteria dominated in all the studied samples. The	Thyme oil in conjunction with MAP 2 provided a shelf life extension of 11 days (shelf life of		Kostaki et al., 2009.



2.	60% CO ₂ /10% O ₂ /30% N ₂ .			respectively, was determined by the use of TVBN as the acceptability parameter (10 mg N/100 g the limit). Similar results were in the case of TMA- N were the acceptability limit (4 mg N/100 g) was exceeded on 6, 9, 9–10, 13, and 19th day, respectively.	acceptability limit of 7 log CFU/g for TVC was exceeded after seven days for control samples while a 2-, 3-, 5- and 12-day life extension was achieved on samples treated with Air+T, MAP 1, MAP 2 and MAP 2+T, respectively.	control samples was six days).	
Sea bass (<i>Dicentrarchus labrax</i>) filets							Provincial et al., 2010.
1.	40% CO ₂ /60% N ₂ .	Polyethylene / polyamide	4 ± 1°C/21 days	No statistically significant differences emerged on TBARS evaluation with the highest values for each studied group to be 0.17 mg MDA per kg at day 11 for 40 / 60 samples, 0.22 mg MDA per kg at day seven for 50 / 50 samples and 0.21 mg MDA per kg for 60 / 40 samples at day 21.	Shelf life of samples stored under 60/40 and 50/50 MAP was doubled while samples under 40/60 was extended by four days with the acceptability limit for APC being set at 6 log CFU/g. Psychrophile load determined shelf life at seven days for 40/60 samples and 14 for 50/50 and 60/40 batches.	High CO ₂ level atmospheres played a key role in preserving sensory characteristics of the treated samples. Drip loss on MAP treated samples was between 4.2 and 5%.	A seven-day shelf life extension (control samples had a shelf life of seven days) was achieved for high CO ₂ MAP samples.
2.	50% CO ₂ /50% N ₂ .	laminated film with OTR: 40–50 cm ³ /m ² /day					
3.	60% CO ₂ /40% N ₂ .						
Aquacultured sea bass (<i>Dicentrarchus labrax</i>).							Reale et al., 2008.
1.	40% CO ₂ /20% O ₂ /40% N ₂ .	The film used was an impermeable PE bag. Samples were submitted to a γ-irradiation of 3 kGy.	2 ± 1°C/10 days.	During the storage period, malondialdehyde values of irradiated and MAP 2 samples (0.25 and 0.29 mg/kg, respectively) remained low while the lowest TBARS levels were recorded in the MAP 2 samples.	Initial APC, PC, <i>Enterobacteriaceae</i> , <i>enterococci</i> and H ₂ S-producing bacteria population was restrained by irradiation to 1 log CFU/g. After seven days, MAP 2 kept LAB, faecal coliforms, <i>enterococci</i> and <i>B. thermosphacta</i> at low levels (3.2, 3.2, and 4.4 log CFU/g, respectively) while irradiation kept their numbers at the level of 1 to 2 log CFU/g.	MAP 1 and 2 sensorial shelf life was extended until the seventh day of storage while irradiated samples were considered unacceptable on the fourth day. Also samples under MAP had a higher rigor index than control and irradiated samples.	
2.	60% CO ₂ /5% O ₂ /35% N ₂ .						
<i>Dicentrarchus labrax</i> filets.							
1.	Vacuum packaging.	For MAP 1 a Cryovac 2050 LID film was used. For MAP 2 a rigid Cryovac tray UBRT 1621 without pad and the same 2050 LID was used.	3°C/26 days.	For MAP B and steam cooking filets putrescine, cadaverine, spermidine and spermine values were 9, 5, 15, and 0 and 12, 5, 4, and 4 mg/kg.	Sensory attributes degradation determined shelf life of samples to be 10 days for control and MAP A and 12 days for steam cooked and MAP B.	A two-day shelf life extension was achieved by steam cooking and MAP B.	Mercogliano et al., 2009.
2.	Stream cooking system.						
3.	70% CO ₂ /5% O ₂ /25% N ₂ for MAP A.						

(Continued on next page)

Table 1. (Continued)

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)— shelf life extension.	Refs
	4. 63% CO ₂ /15% O ₂ /22% N ₂ for MAP B			respectively on the 22nd day of storage.		Increase in drip loss, acid odor, yellowish color and loss of texture were the main reasons for rejection.		
Gutted farmed bass (<i>Dicentrarchus labrax</i>).	1. 0% O ₂ /70% CO ₂ . 2. 20% O ₂ /70% CO ₂ . 3. 30% O ₂ /60% CO ₂ . 4. 40% O ₂ /60% CO ₂ . 5. 30% O ₂ /50% CO ₂ . 6. 21% O ₂ /0% CO ₂ .	A PA/EVOH/PE was used with OTR: 1.5 cm ³ /m ² /day.	3 ± 1°C/9 days.	pH fluctuation was smaller on samples stored under MAP 4 and 5.	APC and <i>Enterobacteriaceae</i> reached, respectively, 2.5 · 10 ⁶ CFU/g and 5.2 · 10 ⁴ CFU/g for MAP 2 and 2.2 · 10 ⁷ CFU/g and 8.0 · 10 ⁵ CFU/g for MAP 5 on the ninth day of storage.	Judging by the sensory characteristics of the samples, MAP 2 and MAP 5 had the best results.	MAP 5 was the best one to preserve the quality of the gutted farmed bass.	Torrieri et al., 2006.
European sea bass (<i>Dicentrarchus labrax</i>) filets.	1. 40% CO ₂ /60% N ₂ . 2. Air. The fish/gas ratio was 1:1.5.	Filets were cut at the beginning or were prepared from the whole ungutted fish stored in ice (ROUND).	2 ± 1°C/8 days.	MAP filets had the highest MDA levels (0.329 mg/kg).	A high-level correlation between <i>Streptococcus spp.</i> growth and odor scores suggests that is the main spoilage organism on the applied MAP. TVC was restrained under MAP and ROUND treatments (5.6, 5.8, and 8.6 log CFU/g for ROUND, AIR and MAP respectively). <i>Pseudomonas spp.</i> and <i>S. putrefaciens</i> were inhibited more severely on MAP samples compared to <i>Streptococcus spp.</i> and <i>Lactobacillus spp.</i> (4.58, 5.74, 5.45, and 5.26 log CFU/g, respectively, on the eighth day of storage).	ROUND filets preserved their sensory attributes better than the other samples from the fifth to the eight storage day.	ROUND filets had the longest shelf life (10, 8, and seven days after slaughtering for ROUND, MAP and AIR samples, respectively).	Poli et al., 2006.
Sea bass slices	1. 60% CO ₂ /40% N ₂ . 2. 60% CO ₂ /10% O ₂ /30% N ₂ . 3. 60% CO ₂ /20% O ₂ /20% N ₂ . 4. 80% CO ₂ /20% N ₂ .	A Cryovac film with OTR: 3–6 cm ³ /m ² /day.	4°C/21 days	High CO ₂ concentrations restrained TVB, TMA, ammonia and formaldehyde formation (0.19, 0.04, 0.16, and 0.35 mg/g, respectively, for samples stored under 100% CO ₂).	The lowest levels of APC and LAB were detected on 100% CO ₂ MAP (5.9 and 6.0 log CFU/g for APC and LAB, respectively, at the end of the storage period).	Sensory attributes of samples stored under MAP with 80 and 100% CO ₂ were considered acceptable for 21 days.	The use of atmospheres rich in CO ₂ (80–100% CO ₂) could lead to a shelf life extension of 12 days (control samples had a	Masniyom et al., 2002.

Study	Product	Storage Conditions	Shelf Life (days)	Findings
5. 80% CO ₂ /10% O ₂ / 10% N ₂ 6. 80% CO ₂ /20% O ₂ 7. 100% CO ₂ .The fish/gas ratio was 1:2 (v/v).	<i>Lateolabrax japonicus</i> .	1. 40% CO ₂ /30% O ₂ /30% N ₂ Samples were superchilled (30 minutes in—1.5°C brine with 3.3% NaCl) (Sc) prior to packaging, LDPE/PA/LDPE barrier pouches with OTR: 52.2 cm ³ /m ² /day were used.	TVB-N, TBA, pH and myofibril fragmentation index for Sc + MAP (19 mg/100 g, 1.45 mg MDA/kg, 6.65 and 135 for TVB, TBA pH and MF, respectively) increased in lower rates compared on samples stored under MAP (25 mg/100 g, 1.92 mg MDA/kg, 7.12 and 160 for TVB, TBA, pH and MF, respectively) on the 16th day of storage.	APC and H ₂ S producing bacterial populations of pretreated MAP samples (Sc + MAP) were lower than that of MAP ones (5.89 and 6.41 and 6.47 and 6.91 log CFU/g for TVC and H ₂ S forming bacteria, respectively, on the 16th day of storage).
80% CO ₂ /10% O ₂ /10% N ₂ . The fish/gas ratio was 1:3.	Sea bass slices	Film with OTR: 46.6 cm ³ /m ² /day. Treatment with Sodium Pyrophosphate. <i>Listeria monocytogenes</i> and <i>Escherichia coli</i> O157 were inoculated on sea bass slices	4°C/21 days	PP pretreated samples stored under MAP had the lowest microbial counts (5.6 and 3.8 log CFU/g for APC and LAB, respectively), compared to MAP (6 and 4.95 log CFU/g, respectively) and control samples (7.3 and 4.2 log CFU/g, respectively). Lag phase on <i>L. monocytogenes</i> was prolonged and growth rate of <i>E. coli</i> O157 was minimized by the use of PP pretreatment.
1. Air 2. Vacuum 3. 70% CO ₂ /30% N ₂ .	Smoked sea bass	Samples were salted with 100% NaCl or 50% NaCl/50% KCl and subsequently were smoked.	4°C/42 days.	Textural parameters and color attributes were similar on samples salted with both methods.
				The use of potassium as a salting agent can effectively reduce sodium content for consumption.

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Table 1. (Continued)

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)— shelf life extension.	Refs
Smoked sea bass	1. Air 2. Vacuum 3. 70% CO ₂ /30% N ₂ .	Samples were salted with 100% NaCl (Na) or 50% NaCl/ 50% KCl (Na-K) and subsequently were smoked.	4°C/42 days.	increased water holding capacity and decreased pH changes. TVBN, TMA and TBA values were similar in both methods used (30– 35 mg/100 g, 18.5– 20 mg/100 g and 0.3– 0.45 mg MDA/kg for TVB-N, TMA and TBA, respectively). On the other hand, biogenic amine formation was delayed on samples salted with NaCl-KCl mixture (3.1, 22.1, and 6 and 28.1, 38.2, and 30.4 for histamine, putrescine and cadaverine on samples stored under MAP and salted with Na-K or Na, respectively, on the 42nd day of storage).	MAP and VP effectively suppressed microbial degradation of smoked sea bass compared to control (5.5–6 and 7.3 log CFU/g for mesophiles, 4.2–4.8 and 5.6 log CFU/g for yeasts and moulds, and 5.5–5.9 and 7.3 log CFU/g for samples stored under MAP-VP salted with both methods and control samples, respectively).		The shelf life of smoked sea bass under both salting methods and storage techniques is under 28 days. MAP and VP showed similar results in preventing spoilage.	Fuentes et al., 2011.



Table 2. Analysis of storage conditions of mussels under various conditions and their effect on microbiological, sensory and chemical attributes.

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)—shelf life extension.	Refs
Mussels (<i>Mytilus galloprovincialis</i>).	1. 60% CO ₂ /20% O ₂ /20% N ₂ . 2. 60% CO ₂ /40% N ₂ . 3. Vacuum packaging.	A PA/LDPE film was used 100 µm thick with OTR: 82 cm ³ /m ² /day.	6°C/14 days.	Both TVBN and TMAN levels were lower in MAP 1 following the order for TVBN: MAP 1 (36 mg/100 g) < MAP 2 (45 mg/100 g). VP (44 mg/100 g) < air (control) samples and: MAP 1(10.2 mg/100 g) < air < MAP 2 (15.2 mg/100 g) < VP whereas TBA values of MAP 1 (1.4 mg/kg) and air samples were significantly higher than the TBA values of VP (0.72 mg/kg) and MAP 2 (0.97 mg/kg) samples.	MAP 1 (6.9 log CFU/gr) had the lowest microbial counts compared to all the other treatments.	MAP 1 samples demonstrated a shelf life of 10–11 days. MAP 2 and VP up to 7–8 days while control samples up to—five to six days of storage based on odor scores.		Goulas 2008.
Live mussels (<i>Mytilus galloprovincialis</i>).	1. 20% CO ₂ /5% O ₂ /75% N ₂ . 2. 50% CO ₂ /20% O ₂ /30% N ₂ . 3. 50% O ₂ /50% N ₂ . 4. 75% O ₂ /25% N ₂ .		2–3°C/6 days.	TVB values for samples under MAP 4 fluctuated between 8.47 mg and 10.42 mg TVB-N/100 g for days one and six, respectively.	Atmospheres with high CO ₂ concentrations negatively affected mussels' survival leading to mortality rate of 93% and 95% for MAP 1 and 2, respectively, after six days. On the other hand presence of high O ₂ levels reduced mortality rates. In samples 3 and 4, the mortality was 17% and 5% after four days and 33% and 10% after six days.	Sensory characteristics of samples stored under MAP 4 while cooked were optimal throughout the whole storage period	The high O ₂ atmosphere (MAP 4) offered a shelf life extension of two to three days compared to commercial storage.	Pastoriza et al., 2004.

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Table 2. (Continued)

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)—shelf life extension.	Refs
Mussels (<i>Mytilus galloprovincialis</i>).	1. 50% CO ₂ /50% N ₂ . 2. 80% CO ₂ /20% N ₂ . 3. 40% CO ₂ /30% O ₂ /30% N ₂ . 4. Vacuum packaging.	LDPE/PA/LDPE barrier pouches were used with OTR: 52.2 cm ³ /m ² /day.	4 ± 0.5°C/15 days.	TVBN and TMA levels detected on control and VP samples exceeded the proposed limit of 35 mg N/100 g and 12 mg N/100 g, respectively, after 15 days of storage. MAP treated samples were acceptable for the whole 15 days. TBA limits of 1 mg MDA/kg were not exceeded with the exception of control samples.	TVC was reduced by 0.9–1.0 log CFU/g. <i>Pseudomonas</i> spp. by 0.7–0.8 log CFU/g, LAB by 1.0–2.2, H ₂ S-producing bacteria by 0.7–1.2. <i>Enterobacteriaceae</i> were not significantly affected by MAP conditions.	Based on odour and taste evaluation, MAP 1 and MAP 3 stored samples had a shelf life 11–12 days. MAP 2 had a shelf life of 15 days while the VP and control samples remained acceptable until day 10–11 and 8–9 of storage, respectively.	MAP 2 gas mixture was the most effective for mussel preservation achieving a shelf-life of 14–15 days. A shelf-life extension of refrigerated mussels by five to six days under MAP may be obtained.	Goulas et al., 2005.
Mussels (<i>Mytilus galloprovincialis</i>).	1. 50% CO ₂ /50% N ₂ . 2. 80% CO ₂ /20% N ₂ . 3. 65% CO ₂ /35% N ₂ . 4. Vacuum packaging.		2 ± 1°C/12 days.	TVB-N and TMA-N values of MAP 2 remained lower than the proposed acceptability limits of 35 mg N/100 g and 8 mg N/100 g, respectively, up to eight days of storage.	MAP 2 and MAP 3 delayed microbial growth compared to MAP 1. After the storage period of 12 days, total viable count of VP, MAP 1, MAP 2, and MAP 3 increased to 8.48, 7.20, 6.80, 6.94 log CFU/gr. After eight days storage, PC had increased from 3.24, 3.01, 2.92, 2.28, 2.89 to 8.47, 6.06, 5.23, 4.76, 4.92 log CFU/g for air, VP, MAP 1, MAP 2, MAP 3, respectively.	Based on microbiological and chemical analyses along with sensory evaluation, MAP 2 and MAP 3 gave a longer shelf-life compared with VP and MAP 1. MAP 2 gas mixture was the most effective for mussel preservation.		Caglak et al., 2008.
Live Mediterranean mussels (<i>Mytilus galloprovincialis</i>).	A. Air. B. 75% O ₂ /25% N ₂ . C. 85% O ₂ /15% N ₂ .	A PS-EVOH-PE barrier film was used. The mussels were of different sizes (30 and 44 units/kg—batch 1 and 2, respectively).	2 ± 1°C/14 days.	On the 10th day of storage no glycogen consumption was evident in all examined samples. The production of ammonium and volatile fatty acids was more intense		A mortality rate of 20% was set as the acceptability limit for shelf life designation. The limit was reached for the mussels of batch 1 packaged with atmospheres A, B and C on day	A high O ₂ atmosphere (75–85% O ₂) is optimal for preservation of high quality live mussels for a storage period of eight days.	Bernardez and Pastoriza, 2011.



				in smaller mussels, which decreased as initial oxygen increased. The values of volatile acids (VFA) reached on the last day of storage (day 14) were 0.709 ± 0.042 for A, 0.256 ± 0.005 for B and 0.22 ± 0.034 for the mussels packaged in atmosphere C.	8, 13, and 13, respectively. In batch 2, this limit was reached before day eight for A, before day 13 for B and on day 13 storage for C.	
Mussels (<i>Mytilus edulis</i>)	65% O ₂ / 35% N ₂ .	A barrier film was used. Samples were inoculated with 6 strains of <i>C. botulinum</i> .	3 and 12°C for 13 and 21 days.	No toxin production was detected even at abusive temperatures or beyond the shelf life limit.		Newell et al., 2012.
Mussels (<i>M. galloprovincialis</i>)	Various levels of CO ₂ were used for cooked samples (20, 55, and 90%) with addition of nisin (21, 115, and 210 IU/g). For live samples storage atmospheres were 30, 60, and 90% O ₂ .	HDPE bags were used. Samples were inoculated with <i>L. monocytogenes</i> biofilm adapted to benzalkonium chloride	2.5°C for 20 days for cooked samples and 12 days for live samples.	<i>L. monocytogenes</i> strains resistant to benzalkonium chloride showed increased resistance to both nisin and MAP compared to non-adopted samples. On live samples, <i>L. monocytogenes</i> populations were not effectively restrained by O ₂ enriched atmospheres.		Ibusquiza et al., 2011.
Live Mediterranean mussels (<i>Mytilus galloprovincialis</i>).	A. Air. B. 20% O ₂ /80% N ₂ . C. 83% O ₂ /17% N ₂ .	A PS-EVOH-PE barrier film was used. The mussels were of different sizes (52 and 41 units/kg).	$2 \pm 1 - 7 \pm 1^\circ\text{C}/10$ days.	Volatile fatty acids detected on the interval fluid of samples stored under MAP C were half the amount detected on samples under MAP B. Lower concentrations were detected on storage at lower	TVBN values and organoleptic characteristics score were inversely related with the latter deteriorating with temperature increase.	Bernandez and Pastoriza, 2013.
				The lowest microbial counts were detected on medium sized samples stored under MAP C at 2°C (3.85 and 2.56 log CFU/g for TVC and H ₂ S producing bacteria, respectively, on day nine).	High-quality products were preserved under MAP B at 2°C for 9 days of storage, resulting to a shelf life extension of five to six days compared to control.	

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Table 2. (Continued)

Species	Initial gas mix	Reference material— Pretreatment	Storage temperature (°C) and storage period (days)	Chemical Parameters— Weight loss.	Microflora	Sensory analysis	Shelf life (days)—shelf life extension.	Refs
Stuffed mussel	A. 50% CO ₂ / 50% N ₂ . B. 100% CO ₂ .		4°C.	temperatures. TVB-N on day eight on mussels stored under high O ₂ atmospheres at 2°C was 16.9 and 15.3 mg/100 g for small and medium sized samples, respectively.	Microbial counts of mesophiles and psychrophiles were lower on MAP treated samples (5.54, 5.51, and 4.54 for TVC and 4.75, 4, and 4 log CFU/g for psychrophiles of control, MAP A and MAP B samples, respectively, on the 13th day).	MAP A storage led to improved sensory characteristics compared to both MAP B and control samples.	A shelf life extension of four days (7 and 11 was the shelf life of control and MAP treated samples, respectively) was achieved with both MAP treatments.	Ulusoy and Ozden, 2011.
				TVBN values were significantly lower on MAP samples (21.49, 21.50, and 28.5 for MAP A, MAP B and control samples, respectively, on the 13th day).				

A high CO₂ atmosphere (80% CO₂/10% O₂/10% N₂ for MAP 3) was opted for as the best for preserving green mussels at 4°C among four atmosphere modifications (40% CO₂/10% O₂/50% N₂ for MAP 1, 60% CO₂/10% O₂/30% N₂ for MAP 2 and 100% CO₂ for MAP 4). On the 15th day microorganisms (6.6 and 6.2 for TVC and 5.6 and 5.2 log CFU/g for LAB on samples under MAP 3 and MAP 4, respectively) and chemical quality (0.052 and 0.048 for TMA and 0.24 and 0.23 mg/kg for TVB on samples under MAP 3 and MAP 4, respectively) were better preserved under MAP 3 and MAP 4 (Masniyom et al., 2011) (Table 2).

Trout

Rainbow trouts were fed with two types of food astaxanthin and canthaxanthin, slaughtered and stored under MAP (40% CO₂/60% N₂ for MAP 1 and 60% Ar/40% CO₂ for MAP 2) at 2°C for 26 days. TBARS concentration was smaller in samples stored under Ar atmosphere compared to that of high CO₂. The inclusion of Argon led to doubling the shelf life of investigated samples (Choubert et al., 2008).

405 previously phenotyped LAB originating from spoiled salted, vacuum-packaged, sodium and potassium nitrite-treated and cold-smoked rainbow trout that were stored at 4 and 8°C were characterized using ribotyping. *Leuconostoc mesenteroides* subsp. *mesenteroides* displayed the highest relative proportion on all the samples stored at 4°C, while *Leuconostoc citreum* was detected mainly in the samples stored at 8°C and, particularly, in samples treated with potassium and sodium nitrite (Lyhs et al., 1999).

The method of hot smoking combined with MAP (10% CO₂/90% N₂ for MAP 1 and nitrogen flush for 30 seconds and VP for MAP 2) or VP was tested on trout inoculated with *L. monocytogenes* stored at 3 and 7°C for 30 days. Increase in storage temperature resulted in greater microbial growth whereas, at 3°C inoculated *Listeria* remained close to the initial numbers (2.3 log CFU/g for MAP 1 samples at the end of storage). Hot smoked samples remained acceptable in terms of sensory characteristics up to 30 days at both temperatures (Shin et al., 2008).

Dufresne et al. (2000) investigated the ability of *C. botulinum* strains to produce toxins on rainbow trouts fillets stored under various MA (100% CO₂ for MAP 1, 25% O₂/75% CO₂ for MAP 2, 50% O₂/50% CO₂ for MAP 3 75% O₂/25% CO₂ for MAP 4 and 100% O₂ for MAP 5) at 12°C for six days. All samples turned toxic by day five though sensory spoilage had already preceded.

A research conducted by Choubert et al. (2005) monitored the effect of light exposure and VP on slices of smoked rainbow trout fed with astaxanthin stored for 15 days. TBARS values were higher in samples exposed to light during storage. Fillets under light also became paler compared to samples stored in the dark. VP was found to have minimal effect on astaxanthin content.

The use of Argon on MAP technology was investigated by Randell et al. (1997) by using various atmosphere modifications (35% CO₂/32.5% Ar/32.5% N₂ for MAP 1, 35% CO₂/65% Ar for MAP 2 and 40% CO₂/60% N₂ for MAP 3) or vacuum for the packaging of rainbow trout fillets at 2°C. The use of MAP 1 and MAP 3 led to reduced microbial numbers at the end of the 11-day storage period (2.7 and 5.6 log CFU/g for coliform and

APC, for both MAP 1 and MAP 3, respectively). Gas packaging was shown to slightly extend the shelf life of fillets compared to vacuum packaging (Fig. 4).

Three different gas mixtures (100% CO₂ for MAP 1, 2.5% O₂/90% CO₂/7.5% N₂ for MAP 2, 30% O₂/40% CO₂/30% N₂ for MAP 3) were tested in an attempt to maintain the microbial and sensory quality of rainbow trout fillets stored at 4 ± 1°C. Mesophiles were limited to 5 log CFU/g on the 14th day while psychrophiles reached 7 log CFU/g on the 10th day of storage. *Enterobacteriaceae* detected on samples under MAP 1 were 0.5 to 1.5 log units lower compared to MAP 2 and MAP 3, respectively. MAP 1 and MAP 2 maintained TVB-N values under the limit of 25 mg/100 g until day 12 (Arashisar et al., 2004) (Fig. 5).

Hot smoked rainbow trout fillets were inoculated with *L. monocytogenes* strains and stored under vacuum and two MAs (100% CO₂ for MAP 1 and 50% CO₂/50% N₂ for MAP 2) at 2°C for seven weeks. The microbiostatic effect of carbon dioxide was evident in MAP 1 displaying the best results in inhibiting *Listeria* growth. MAP 1 and VP extended shelf life up to four weeks while MAP 2 samples were acceptable for one more week (Erkan et al., 2009).

The impact of slaughtering method (percussive stunning and death in ice slurry) on the quality parameters of rainbow trout stored under MAP (30% O₂/40% CO₂/30% N₂) at 2 ± 2°C for 18 days was investigated by Ozogul and Ozogul (2004). K value was significantly lower in samples stored under MAP and killed with the percussive stunning method. Slaughtering by percussive stunning proved to be more effective in quality retention compared to the ice slurry method.

The potential of MAP (60% CO₂/40% N₂ for MAP 1 and 80% CO₂/20% N₂ for MAP 2) to limit the growth of *Aeromonas* spp. and *Yersinia enterocolitica* inoculated on trout fillets stored at 0 and 5°C for 21 days and 12°C for 14 days was evaluated by Davies and Slade, (1995). The growth of the two pathogens was greatly affected by both the atmosphere composition and storage temperature (3.5 and 3.7 log CFU/g at 0°C, 7 and 4.9 log CFU/g at 5°C, and 7.3 and 7.5 log CFU/g at 12°C for *Aeromonas* stored at MAP 1 and 2, respectively) (6.6 and 5.3 log CFU/g at 0°C, 8.2 and 7.5 log CFU/g at 5°C, and 8.1 and 8 log CFU/g at 12°C for *Yersinia* stored at MAP 1 and 2, respectively).

Sugar-salted trout fillets were VP and studied versus storage at 3 and 8°C until spoilage (27 and 21 days, respectively). Mesophilic and psychrophilic populations were 1 to 2 log CFU/g lower in samples stored at 3 compared to those stored at 8°C. A two-day shelf life extension of fillets stored at 3°C was reported compared to the 18-day shelf life of samples at 8°C based on sensory and microbiological analyses (Lyhs et al., 2001).

The effectiveness of MAP (10% O₂/50% CO₂/40% N₂ for MAP 1, 10% O₂/50% CO₂/40% Ar for MAP 2, 20% O₂/50% CO₂/30% N₂ for MAP 3, 20% O₂/50% CO₂/30% Ar for MAP 4, 30% O₂/50% CO₂/20% N₂ for MAP 5 and 30% O₂/50% CO₂/20% Ar for MAP 6) and VP on the preservation of filleted rainbow trout was studied by Gimenez et al. (2002). Samples stored under low oxygen atmospheres (MAP 1 and 2) had lower lipid oxidation (2.5–3 mg MDA/kg), and TVB level did not exceed the acceptability limit (250 mg N/kg) during the whole storage period of 20 days at 1 ± 1°C.

A decrease in astaxanthin and canthaxanthin concentration after 11 days of storage of rainbow trout fed with

ketocarotenoids and stored under vacuum at 4°C was recorded when TBARS values reached their maximum values that is 5.9 nmol of TMP/mg of lipid. Based on microbiological criteria (6 log CFU/g of TVC is the acceptability limit), the shelf life of samples was limited down to five days (2 and 5.6 log CFU/g were the psychrotrophs and coliforms counts, respectively, on day five) (Gobantes et al., 1998).

The detection of spoilage flora on cold-smoked rainbow trout fillets packaged under vacuum, treated with nitrite and nitrate and stored at 4 and 8°C was studied by Lyhs et al. (1998). LAB were the predominant species with 76% of the 620 isolates detected. Eighty five of the isolates belonged to the *Enterobacteriaceae* species. For samples stored at 8°C and treated with nitrate the incidence of *Pseudomonas aeruginosa* and staphylococci were shown to pose a risk for public health.

A shelf life of more than six months was reported for gravad (fillets rubbed with 1:2 mixture of salt and sugar in the amount of 350 g/kg of raw material, and matured for 48 h at 3°C) rainbow trout stored under vacuum at -30°C while vacuum storage proved to be more effective than MAP (25% O₂/60% CO₂/15% N₂ for MAP 1 and 40% N₂/60% CO₂ for MAP 2) prolonging the shelf life up to eight weeks at 3°C (Michalczyk et al., 2008).

The role of oregano essential oil (0.2 for M₁ and 0.4% v/v for M₂) as a “natural” preservative on fresh, salted rainbow trout fillets stored under MAP (5% O₂/45% CO₂/50% N₂) at 4°C for 21 days was examined by Pyrgotou et al. (2010). The addition of oregano oil limited microbial growth (7.2 and 5.7, 5.3 and 4.5, 4.59 and 4.33, 5.7 and 5.6, 5.4 and 4.9 log CFU/g for TVC, H₂S producing bacteria, *Pseudomonads*, LAB, and *Enterobacteriaceae* on M₁ and M₂ samples, respectively) and retained TVB-N and TMAN values (25.23 and 21.2 and 2.65 and 2.06 mg/100 g for TVB-N and TMAN on M₁ and M₂ samples, respectively). The shelf life extension of M₁ samples amounted to seven to eight days, whereas the release of strong odors limited shelf life of M₂ samples (Fig. 6).

Inoculated *L. monocytogenes* on rainbow trout fillets stored under MAP (50% CO₂/50% N₂ for MAP 1, 80% O₂/20% CO₂ for MAP 2 and 2.5% O₂/90% CO₂/7.5% N₂ for MAP 3) at 4 ± 1°C was significantly inhibited (5.3–6.2 log CFU/g for MAP samples on the 18th day) compared to control (7.4 log CFU/g) samples. The highest TVB-N content was detected on the control samples (27–31.5 for MAP samples and 50 mg/100 g for control samples) (Yilmaz et al., 2009).

VP smoked rainbow trout originating from 6 smokehouses was examined after three weeks storage at 2 ± 1°C. Microbial numbers varied between 4.81 and 8.25 log CFU/g for LAB, 1.71 to 2.25 log CFU/g for psychrotrophic clostridia, 1.36 to 4.48 for *Enterobacteriaceae*, 2.78 to 4.33 for fungi and 2.75 to 4.81 for *Aeromonas* while *Salmonella*, *Escherichia coli*, and *Listeria monocytogenes* were not detected (Gonzalez-Rodriguez et al., 2002).

Wolf-fish

Superchilling conditions (-1°C) were applied on farmed spotted wolf-fish packaged under MA conditions (60% CO₂/40% N₂). The combination of those two preservation methods limited psychrotrophs growth (6.5 log CFU/g on day 15) while samples displayed the lowest TMA and TVBN values on day 15

(2 and 13 mg/100 g). Under these storage conditions, shelf life extended to 15 days compared to 8–10 days for superchilled control samples. However, for samples stored under MAP and air at 4°C the shelf life was 13 and six to eight days, respectively (Rosnes et al., 2006).

Salmon

Schirmer et al. (2009) investigated the effect of MAP (100% CO₂ with gas/product ratio 0.2/1.0 v/v) and brine solution containing several contents of citric acid (3% w/w, pH 5), acetic acid (1% w/w, pH 5) and cinnamaldehyde (200 µg/mL) on salmon samples stored at 4°C. Combination of pretreatment with MAP restrained microbial populations of both inoculated and natural strains (2.5, 2.5, and 0.7 log CFU/g for TPC, LAB and *Enterobacteriaceae*, respectively on samples pretreated with citric and acetic acid) for 14 days.

Detection of volatile compounds in king salmon stored under MAP (40% CO₂/60% N₂) led to the conclusion that several alcohols (cyclopentanol, Z-2-penten-1-ol, 1-penten-3-ol, and 1-octen-3-ol) and aldehydes (hexanal, octanal, E-2-pentenal, and E-2-hexenal) can be characterized as freshness indicators. It is noteworthy that acetoin, ethyl benzene, propyl benzene, styrene, 3-methyl butanoic acid and acetic acid were detected on spoiled samples. The use of anesthetic containing isoeugenol led to increase of E- and Z-isoeugenol levels (Wierda et al., 2006).

For cold-smoked salmon fillets stored under vacuum (UV) an increase in storage temperature resulted in shelf life reduction (for samples at 0, 2, 4, 6, and 8°C shelf life was 26, 21, 20, 10, and 7 days, respectively). TVBs amounted to 31.8, 29.3, 29.8, 30.0, and 29.9 mg TVB-N/100 g at 0, 2, 4, 6, and 8°C at the end of their storage life, respectively, while TMA was 10.2, 7.3, 7.5, 7.4, and 7.7 mg TMA-N/100 g at 0, 2, 4, 6, and 8°C after 26, 21, 20, 10, and 7 days, respectively (Dondero et al., 2004).

Fagan et al. (2004) after having stored freeze-chilled (-35°C for 2.5 hours and -30°C for three days and subsequent storage at 2–4°C) salmon fillets under MAP (40% CO₂/60% N₂) for seven days concluded that the combination of the preservation techniques led to a shelf life extension of two days. On the rejection day (seventh day of storage), MAP samples had 5.71 log CFU/g TVC, 18 mg N/100 g TVB-N and 1.01% FFA (Fig. 1).

High pressure processing (150 MPa for 10 minutes at 5°C) of salmon fillets prior to storage extended the shelf life by two days, while combined with MAP (50% CO₂/50% N₂) led to a prolongation of four days. Even though HP treatment reduced microbial counts (the upper acceptance limit of 7–7.2 log CFU/g was reached after 18 days for HP treated MAP samples), there was a significant downgrading of the color of the samples (Amanatidou et al., 2000).

A significant reduction in microbial growth was obtained with superchilled (-2°C) storage of salmon fillets under MAP (60% CO₂/40% N₂) (1.8, 1 and 2.2 log CFU/g for TVC, H₂S-producing bacteria and psychrotrophs, respectively) resulting in a shelf life two times longer (21 days) than chilled MAP samples (10 days) stored at 4°C and 3 times than control samples (7 days) (Sivertsvik et al., 2003).

The level of inhibition of *L. monocytogenes* growth caused by salting and cold smoking of salmon fillets stored under vacuum

(UV) at 4 and 8°C and the identification of a model that would fit in the process was investigated by Cornu et al. (2006). A secondary model coined by Devlieghere et al. (2001) and modified by Gimenez and Dalgaard (2004) appeared to be the most appropriate one.

The occurrence of LAB and Gram-negative bacteria at high levels on spoiled cold-smoked salmon was investigated by Paludan-Muller et al. (1998). The addition of nisin in conjunction with storage under MAP (60% CO₂/40% N₂) led to shelf life extension of two weeks and a reduction of LAB numbers by 2 logs, while Gram⁽⁻⁾ bacteria were also inhibited (1.7 log CFU/g after seven weeks of storage). *Carnobacterium piscicola* was the strain detected in 87% of the 255 LAB isolates.

Real time PCR revealed that the major groups of bacteria detected on salmon stored under MAP (60% CO₂/40% N₂) at 1 and 5°C for 18 days were *Brochothrix* spp. and *Carnobacterium* spp. Moreover, *B. thermosphacta* and *C. piscicola* were the main isolates identified (Rudi et al., 2004).

The comparison of microbiological parameters affecting fresh and thawed salmon fillets stored under MAP (60% CO₂/40% N₂) at 2°C was conducted by Emborg et al. (2002). The dominant strain isolated from fresh salmon was *Photobacterium phosphoreum* limiting shelf life down to 14 days. Freezing prior to packaging eliminated *P. phosphoreum* growth thereby allowing a shelf life extension by one to two weeks under MAP and enabling the dominance of *Carnobacterium piscicola*. On the 14th day levels of biogenic amines were considerably higher at fresh samples (11.3, 17.5, 37.8, and 17.1 mg/kg for histamine, tyramine, cadaverine, and agmatine, respectively) (Fig. 3).

The application of different levels of CO₂ (0, 10, 20, 30, 40, 60, 80, and 100 cm³) for preserving salmon stored at 0°C for up to 90 days was tested by Fletcher et al. (2005). Usage of higher levels of CO₂ led to poorer overall quality. The levels of hypoxanthine were negatively affected with CO₂ increase. TVB-N values remained under 20 mg/100 g for whole 90 days under all MAs.

A prototype gas-sensor array system was used for the evaluation of sensory characteristics of smoked salmon coming from 4 different smokehouses and stored UV up to four weeks at 5 and 10°C. Both TVC and LAB of the investigated samples varied between 4.1–7.5 and 2.9–4.6 log CFU/g and 4.1–7.5 and 2.7–5.1 log CFU/g for samples at 5 and 10°C, respectively, after 10 and 28 days of storage (Olafsdottir et al., 2005).

Pre-rigor salmon fillets were stored under MAP (60% CO₂/40% N₂) with eight different methods with varying tray sizes (280 and 1800 mL), gas/product ratios (1:1 and 1:2), number of fillet layers and capacities of the CO₂ emitters being used (based on the fish weight or surface area of the fish) at 2°C for 21 days. The quantity of CO₂ affected substantially the microbial load since enhancing amounts resulted in lower microbial counts (3.5 and 5.5 log CFU/g, on the 21st day for low and high CO₂, respectively) (Hansen et al., 2009a).

Pre-rigor salmon fillets were stored either under MAP (60% CO₂/40% N₂) with a gas to product volume ratio (G/P ratio) of 3/1 (traditional MAP) and 1/1 (packaged with a CO₂ emitter) or vacuum at 1.2°C for 25 days. The microbial counts of samples stored under MAP were lower (5.6, 7.2 and 7.9 log CFU/g for TVC under MAP with emitter, MAP and vacuum, respectively on day 25), with *P. phosphoreum* being the dominating strain. Sensory attributes were found to be unacceptable on day

8 and 15 for vacuum and MAP samples, respectively (Hansen et al., 2009b) (Fig. 2).

The optimal storage conditions with alterations of parameters such as MAP conditions (25% CO₂/75% N₂, 40% CO₂/60% N₂, 60% CO₂/40% N₂/75% CO₂/25% N₂, 90% CO₂/10% N₂), g/p ratio, the use of natural additives [rosemary extract (1 g/L) and Sea-i (55 g/L)], superchilling and subsequent storage at 2 ± 2°C for 28 days, were tested by Fernandez et al. (2009). The conditions leading to the best results were 90% CO₂/10% N₂ and g/p ratio of 2.5 resulting in shelf life of 22 days with the microbiological attributes being the limiting parameters. The addition of additives was not found to prolong the shelf life of the samples.

King salmon fillets were subjected to MAP (60% CO₂/40% N₂ and 100% N₂) storage at 0°C for 54 days. The first signs of spoilage were detected on 15, 15, and 21st day for air, N₂ and N₂/CO₂ samples, respectively. Total aerobic counts for MAP samples reached 8 log CFU/g numbers on the last day of the experiment and high microbial numbers were correlated with sensory spoilage. Hypoxanthine levels reached 3.7 µmol/g on day 54 for samples stored under CO₂ atmosphere (Fletcher et al., 2002).

Salmon fed with astaxanthin and canthaxanthin were slaughtered, filleted and stored either raw or smoked UV at –20°C for 12 weeks. Raw fish fed with astaxanthin displayed significant changes after six weeks of storage but not after 12 weeks. However, raw fish with canthaxanthin showed enhanced *L** values, reduced *a** values and carotenoid content decreased from 10.6 down to 4.36 mg/kg. On the contrary, smoked astaxanthin fillets displayed decreased pigment content from 9.39 down to 7.26 mg/kg after 12 weeks (Sheehan et al., 1998).

Two CO₂ enriched atmospheres (20% CO₂/80% air for MAP 1 and 40% CO₂/60% air for MAP 2) were tested for the preservation of salmon steaks stored at 2 ± 1°C for days. MAP 1 and 2 contributed to a shelf life extension of 6 and 15 days, respectively, compared to control samples. On the 22nd day, MAP 2 exhibited the lowest values both for microbial counts (5.9, 5.5, 5.3, and 3.8 log CFU/g for TVC, *B. thermosphacta*, LAB and *Enterobacteriaceae*, respectively) and chemical parameters (12 and 18 mg/100 g for TMA and TVB-N, respectively) (De la Hoz et al., 2000).

The negative effect of temperature abuse (8 and 16°C) on quality and toxin production of *C. botulinum* type E on salmon fillets stored under MAP (75% CO₂/25% N₂) was reported in a research conducted by Reddy et al. (1997b). Toxin detection on samples stored at 8°C occurred simultaneously or slightly after sensory spoilage, while storage at 4°C prevented samples from becoming toxic even after 20 days of sensory spoilage.

Atlantic salmon was bought from a local market and kept under MAP [55% CO₂/45% N₂ for MAP 1 (packs 1) or at MAP 1 for three days and then repacked at 30% CO₂/70% N₂ (MAP 2) for the following 12 days (packs 2)] at 4°C for 15 days after commercial packaging to study its microbial community structure using a culture-based and a DNA-based method. After 15 days in packs 1 the dominant genus was *Carnobacterium* followed by *Lactococcus* and *Vagococcus* sequences while in pack 2 *Shewanella* spp. or *Carnobacterium* spp. proved to be the dominant species (Powell and Tumblin 2012).

Cold-smoked salmon inoculated with *L. monocytogenes* was treated with diacetate and lactate (0 to 0.15% w/w and 0 to

1.5% w/w, respectively) and stored under MAP (40% CO₂/60% N₂) or VP at 8 and 15°C. The addition of diacetate on salmon samples inhibited considerably the growth of *L. monocytogenes* (4.5 log CFU/g lower compared to MAP samples) for more than 40 days at 8°C (Mejlholm and Dalgaard, 2007).

Fernandez et al. (2010) applied various gas mixtures (25, 40, and 75% CO₂ with a g/p ratio of 1.2 and 60, 75 and 90% CO₂ with a g/p ratio of 2.5) and gas to product ratios for the determination of a scale-up parameter for shelf life prolongation of salmon fillets stored at 2 ± 2°C for 28 days. Samples in atmospheres with high CO₂ levels and G/P ratio had a longer shelf life (20–23 days with bacterial count <6 log CFU/g) and with a CO₂ solubility of 1072 to 2065 ppm.

The identification of LAB in salmon stored under MAP (80% O₂/20% N₂ for MAP 1 and 60% N₂/40% CO₂ for MAP 2) at 4°C for six days was attempted by Franzetti et al. (2003). *Leuconostoc spp.* isolates dominated under both MAP 1 and MAP 2.

Smoked salmon VP and stored at 2 ± 1°C was obtained at retail level after three weeks of storage. Microbiological analyses revealed that LAB ranged between 6.26 and 7.59 log CFU/g, psychrotrophic clostridia between 1.71 and 2.08 log CFU/g, *Enterobacteriaceae* between 2.03 and 2.75 log CFU/g while *S. aureus* was detected in 2 of the packages and there was no presence of *Salmonella*, *Escherichia coli*, and *Listeria monocytogenes* (Gonzalez-Rodriguez et al., 2002).

Sea bream

A 99.4% similarity on the 16S rRNA gene sequence between *S. baltica*, *S. putrefaciens* and *S. oneidensis* strains isolated from *Sparus aurata* stored under MAP (40% CO₂/30% N₂/30% O₂) at 0, 10, and 20°C was detected in an experiment conducted by Tryfinopoulou et al. (2007).

Pseudomonas lundensis and *P. fluorescens* were the dominating species in the microbial flora of sea-bream stored under MAP (40% CO₂/30% N₂/30% O₂) at 0, 10, and 20°C. Out of the 106 isolates, proteolytic and less lipolytic strains dominated on MAP samples. Total pseudomonads reached 6 log CFU/g on the MAP samples stored at 2°C after 27 days of storage (Tryfinopoulou et al., 2002).

Sea bream fillets were osmotically treated with 50% high dextrose equivalent maltodextrin (DE 47) plus 5% NaCl with or without the addition of nisin (2 × 10⁴ IU/100 g osmotic solution) stored under MAP (50% CO₂/50% air) at 0, 5, 10, and 15°C. MAP samples were characterized by significantly reduced microbial loads (8.2, 7.8, 5.3, and 7 log CFU/g for TVC, LAB, *B. thermosphacta* and H₂S-producing bacteria after 30 days of storage, respectively). Shelf life of samples osmotically treated, protected with nisin and stored under MAP was prolonged to 48 days compared to 10 days of control samples at 0°C (Tsironi and Taoukis, 2010).

Irradiation (1 and 3 kGy) was tested on salted fillets VP at 4 ± 1°C. Chemical parameters of irradiated samples improved compared to control (6.17, 4.52, and 8.87, and 48.13, 37.21, and 60.52 mg N/100 g for TMA and TVB-N of irradiated with 1 and 3 kGy and control samples, respectively, on the 42nd day). On the other hand, TBA was higher on treated samples (2.15 and 3.26 mg malondialdehyde/kg for 1 and 3 kGy, respectively). The shelf life extension of irradiated salted VP fillets was 13 days (Chouliara et al., 2004).

MAP (40% CO₂/30% N₂/30% O₂) along with the addition of oregano essential oil (0.4 and 0.8% v/w) on lightly salted sea bream fillets stored at 4 ± 1°C led to a shelf life extension of 12–13 days. TVB-N (40.5, 35.2, and 71.8 mg N/100 g for MAP with oregano 0.4 and 0.8% and untreated samples, respectively, on the 33rd day), TMAN (3.25, 3.74, and 7.38 mg N/100 g for MAP with oregano 0.4 and 0.8% and untreated samples, respectively, on the 33rd day) and TBA (0.3, 0.44, and 0.72 mg malondialdehyde/kg for MAP with oregano 0.4 and 0.8% and untreated samples, respectively, on the 33rd day) were effectively controlled on treated samples under MAP (Goulas and Kontominas, 2007).

The evaluation of sensory quality of sea bream stored under MAP (60% CO₂/40% N₂ for MAP 1 and 60% CO₂/10% N₂/30% O₂ for MAP 2) at 2, 4, and 8°C with the use of a quality index method was evaluated by Campus et al. (2011). MAP 1 at 4°C resulted in the maximum storage life of samples (13 days at 4°C). However, further lowering of storage temperature by 2 degrees did not give any beneficial results whereas temperature abuse led to shortening of shelf life by six days.

Eel

Farmed eels were stored either aerobically or under vacuum and MAP (40% CO₂/30% N₂/30% O₂) at 0°C. Their shelf life in terms of sensory attributes was 18 ± 1, 11 ± 1, and 11 ± 1 for MAP, vacuum and control samples, respectively, whereas samples remained acceptable with regard to their microbial parameters (7.1, 7.4, and 7.8; 7.3, 5.9, and 4.2; and 3.4, 3.4, and 3 log CFU/g for TVC, Pseudomonads and *Enterobacteriaceae* of control, vacuum and MAP samples, respectively, with the measurements being performed on the 18th, 31st, and 37th day for control, vacuum and MAP samples, respectively) for 34, 28, and 18 days, respectively (Arkoudelos et al., 2007).

Other farmed fish

Sole

The trial of an innovative foam tray with absorbers for volatile amines and liquids compared with normal trays both under active MAP (40% CO₂/60% N₂) for storage of sole fillets at 3°C was carried out by Franzetti et al. (2001). There was a clear advantage of innovative trays in both microbial (8.69, 8.3, 8.43, and 6.3 and 8.7, 7.25, 7.25, and 6 log CFU/g for TVC, Gram⁽⁻⁾, H₂S-producing bacteria and LAB, for samples in standard and innovative tray, respectively, on the 10th day) and chemical parameters of the samples.

Microbial, biochemical and sensory attributes of sole fish fillets stored under CO₂-enriched atmospheres (20/80 and 40/60 CO₂/air) at 2°C were evaluated by Lopez-Galvez et al. (1998). High CO₂ mixture (40%) caused a delay in microbial growth. The limit of 7 log CFU/g for TVC was reached on the 15th day of storage, with the dominant population being *Br. thermosphacta* (66%) and *Lactobacillus spp.* (33%). No change in pH was detected on samples stored under 40/60 mixture, while TMA and TVBN formation was significantly restrained compared to control (10, 32, and 44 and 22, 38, and 52 mg N/100 g for TMA and TVBN on samples stored under 40/60, 20/80 CO₂ mixtures and control samples, respectively).

Tilapia

Four packaging films (LLDPE/EVOH/LLDPE, 60 μm for MAP 1, ONy/PE 70 μm for MAP 2, OPP/PP 60 μm for MAP 3 and HDPE 50 μm for MAP 4) were tested with the same MA (80% CO_2 /20% N_2) for the preservation of red tilapia at $2 \pm 2^\circ\text{C}$. MAP 1 and MAP 2 provided better conditions for the microbial inhibition (7.8 and 5.9 log CFU/g for TVC and LAB of both MA on day 16) and sensory upkeep giving a shelf life of 14 days for MAP 1 and 2 compared to 9 and 6 days for MAP 3 and 4, respectively (Siah and Mohd Tahir, 2011).

Air storage proved to be more beneficial compared to MAP (50% CO_2 /50% N_2) storage of tilapia fillets under chilled (1°C) and superchilled (-1°C) conditions due to undesirable effect on color of the samples (sixth day). Microbial attributes were better preserved on MAP samples compared to control [13 and 20 and 23 and 23 days for TVC to reach the limit of acceptability (8 log CFU/g) on air and control samples at 1 and -1°C , respectively] (Cyprian et al., 2013).

Fresh tilapia fillets were stored under MAP (75% CO_2 /25% N_2 for MAP A, 50% CO_2 /50% N_2 for MAP B 25% CO_2 /75% N_2 for MAP C) at 4°C into high barrier film (OTR: 5–30 mL/m^2 . day) bags. Shelf life of samples from the sensorial point of view was 9, 13, 20–23, and 27–30 days for control, MAP C, MAB B and MAP A samples, respectively. TMA content remained low in all studied samples (<3 mg/100 g) on the time of spoilage. A serious drawback on MAP C stored samples was the high mesophile load (8.5 log CFU/g) on the day of sensory rejection (27th day) (Reddy et al., 1994).

Shelf life of tilapia fillets stored under MAP (75% CO_2 /25% N_2) at 4°C was extended by more than 12 days compared to control (9 to 13 days) due to microbial growth restraint. Storage at higher temperatures resulted in shorter shelf life (13–16 and 9–13 days for 8 and 16°C , respectively) and higher TMA formation (17.17 and 24.94 for 8 and 16°C , respectively) (Reddy et al., 1995).

Vacuum packaging and MAP (75% CO_2 /25% N_2) were tested as hurdle techniques to avoid toxin production on tilapia fillets inoculated with *C. botulinum* type E and stored at refrigeration (4°C) and abuse temperatures (8 and 16°C). At 4°C toxin detection occurred >10 days after sensory spoilage on both vacuum and MAP stored samples, but on 16°C spoilage and toxin production happened on the same day (third and fourth day for vacuum and MAP samples, respectively) (Reddy et al., 1996).

Catfish

The inoculation of *Listeria ivanovii* was used as a challenge test on smoked catfish stored under MAP (80% CO_2 for MAP 1 and 30% O_2 /40% CO_2 /30% N_2 for MAP 2) at two storage temperatures (2 and 8°C) for smoked catfish. On samples stored under MAP 1 *Listeria* populations were reduced significantly at both temperatures with the same growth pattern followed by TPC (3 and 3.5 log CFU/g at 2 and 8°C , respectively). TBARS of samples stored at MAP 1 was significantly lower compared to MAP 2 and air samples (1.25 and 1.3 mg/kg for samples stored at 2 and 8°C , respectively). The optimal storage conditions of smoked catfish was MAP 1 at 2°C (Goktepe and Moody, 1998).

Reddy et al. (1997a) investigated the shelf life of catfish fillets inoculated with *C. botulinum* type E. strains and stored under MAP (75% CO_2 /25% N_2) at 4, 8, and 16°C for up to 45 days. On all studied samples, sensory spoilage and toxin detection coincided on the same day when stored at 16°C , while at 4°C there was no toxin detection on any of the MAP samples even after 37 days after sensory spoilage. Sensory spoilage came after 38–40, 20–24 and 13 days for MAP, vacuum and control samples, respectively.

Silva and White (1994) studied the effect of in-package CO_2 concentration (25%/75% CO_2 /air for MAP A and 80%/20% CO_2 /air for MAP B) and storage temperature (2 and 8°C) on the attributes of catfish fillets. MAP A samples stored at 2°C had the lowest microbial numbers (5 log CFU/g for APC on

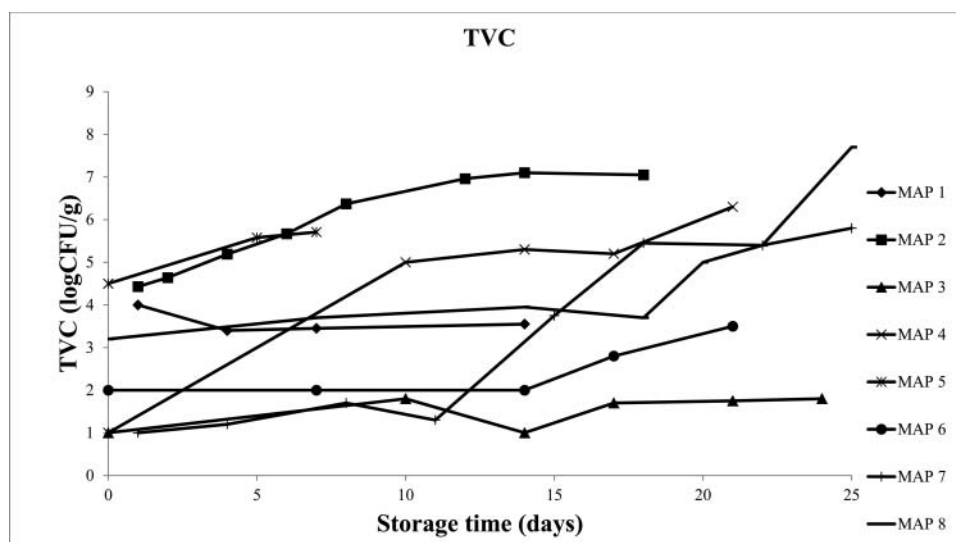


Figure 1. Mesophile growth on salmon samples stored under MAP versus storage time [MAP 1 (100% CO_2 with citric and acetic acid pretreatment) inoculated with *S. putrefaciens*, *Carnobacterium maltaromaticum*, *P. phosphoreum*, and *Yersinia aldovae* at 1:1:1:1 inoculum level, Schirmer et al. (2009), MAP 2 (50% CO_2 /50% N_2) with high pressure pretreatment (150 MPa for 10 minutes at 5°C), Amanatidou et al. (2000), MAP 3 (60% CO_2 /40% N_2) at superchilled storage, Sivertsvik et al. (2003), MAP 4 (60% CO_2 /40% N_2) at chilled storage, Fagan et al. (2004), MAP 5 (40% CO_2 /60% N_2) after freeze-chilling (-35°C for 2.5 hours and -30°C for three days), Fagan et al. (2004), MAP 6 (60% CO_2 /40% N_2 with the ratio of CO_2 emitters and sample weight being 1:1), Hansen et al. (2009b), MAP 7 (60% CO_2 /40% N_2) with gas to product volume ratio 1:1 and the addition of a CO_2 emitter, Hansen et al. (2009) and MAP 8 (90% CO_2 /10% N_2) with a gas to product ratio of 2.5, Fernandez et al. (2009)].

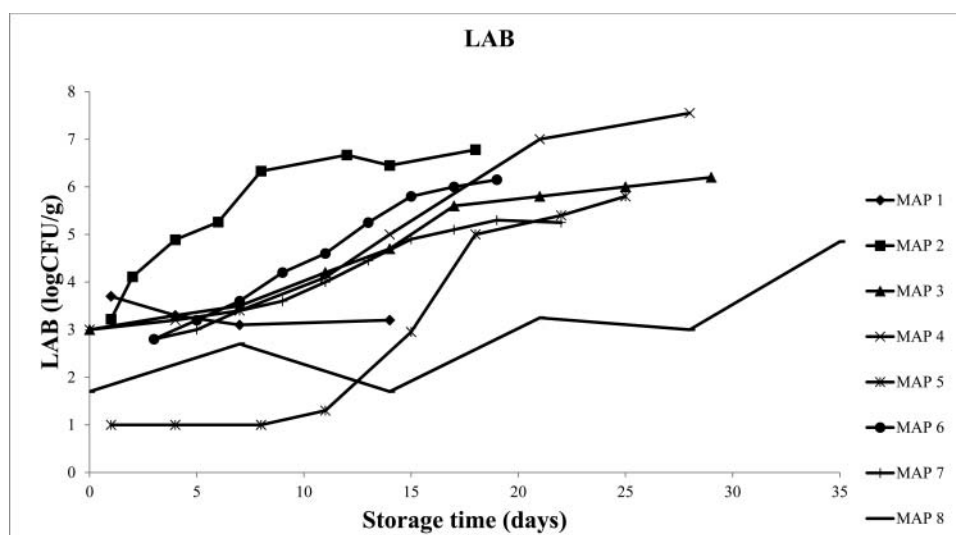


Figure 2. LAB population on salmon stored under modified atmosphere conditions [MAP 1 (100% CO₂ with citric and acetic acid pretreatment) inoculated with *S. putrefaciens*, *Carnobacterium maltaromaticum*, *P. phosphoreum*, and *Yersinia aldovae* at 1:1:1:1 inoculum level, Schirmer et al. (2009), MAP 2 (50% CO₂/50% N₂) with high pressure pretreatment (150 MPa for 10 minutes at 5°C), Amanatidou et al. (2000), MAP 3 (60% CO₂/40% N₂), MAP 4 (60% CO₂/40% N₂) on thawed samples (storage at -20°C for four weeks prior to MAP), Emborg et al. (2002), MAP 5 (60% CO₂/40% N₂) with gas to product volume ratio 1:1 and the addition of a CO₂ emitter, Hansen et al. (2009b), MAP 6 (20% CO₂/80% air), MAP 7 (40% CO₂/60% air), De la Hoz et al. (2000), MAP 8 (60% CO₂/40% N₂) on cold-smoked salmon fillets, Paludan-Müller et al. (1998)].

the third week of storage and no detection of *Salmonella* spp. or *L. monocytogenes*) and proved to be the best studied treatment.

Carp

Carp fillets were stored in several storage conditions [vacuum packaging, MAP 1 (50% CO₂/50% N₂), MAP 2 (30% CO₂/70% N₂), and MAP 3 (80% CO₂/20% N₂)] and at several storage and working room temperatures (3 and 8°C and 5–18°C, respectively). The changes in the working room temperature did not have any significant effect on microbial load of the samples. Storage at 3°C and under MAP 1 atmosphere conditions gave the best results (6.6, 5.3, 3.9, and 6.6 log CFU/g for TPC, *Pseudomonas*, coliforms and LAB on day 21, respectively) and extended shelf life up to 14 days (Marcel et al., 1996).

Fresh carp inoculated with *Salmonella Enteritidis* PT4 and *Listeria monocytogenes* strains was either immersed in a sorbate solution (5%, w/v) or heated (60°C, 1 minutes), or treated with a combination of the two treatments and stored under MAP (30% O₂/40% CO₂/30% N₂) at 0 ± 1°C. The combination of both treatments with MAP was the most effective (4.1 and 3.5, 4.5 and 5.8, 4.2 and 4.6, and 4.3 and 5.1 log CFU/g for *S. Enteritidis* and *L. monocytogenes* treated with combined treatments, hot water, p. sorbate and control samples, respectively, at the 15th day) in restraining microbial growth (Tassou et al., 2004).

An atmosphere modification having CO (5% O₂/25% CO₂/69% N₂/1% CO) was tested for the shelf life extension of carp stored at 2 ± 2°C for 18 days. Loss of sensory quality was evident on the fourth and ninth day for control and MAP samples, respectively. TVBN

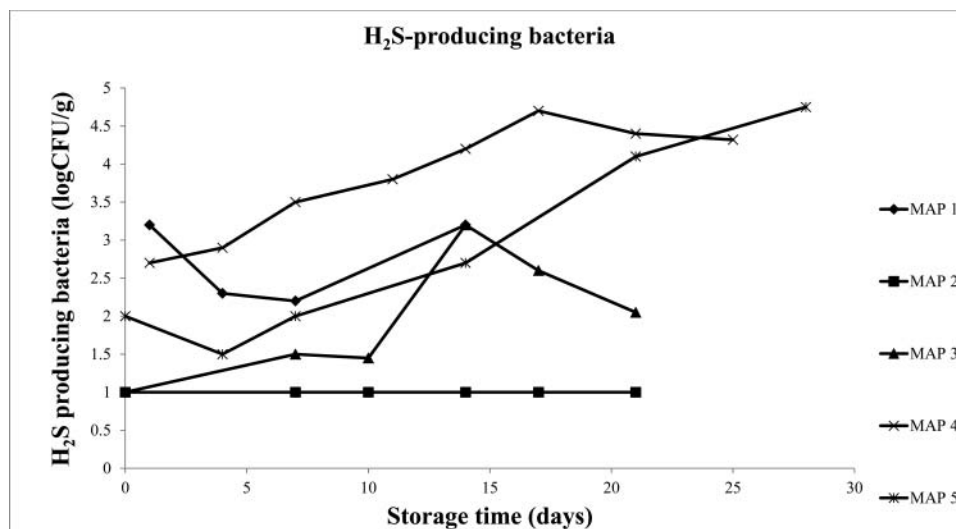


Figure 3. Hydrogen sulphide producing bacteria levels detected on salmon samples stored under MAP [MAP 1 (100% CO₂ with citric and acetic acid pretreatment) inoculated with *S. putrefaciens*, *Carnobacterium maltaromaticum*, *P. phosphoreum* and *Yersinia aldovae* at 1:1:1:1 inoculum level, Schirmer et al. (2009), MAP 2 (60% CO₂/40% N₂) at superchilled storage, MAP 3 (60% CO₂/40% N₂) at chilled storage, Sivertsvik et al. (2003), MAP 4 (60% CO₂/40% N₂) and MAP 5 (60% CO₂/40% N₂) on thawed samples (storage at -20°C for four weeks prior to MAP), Emborg et al. (2002)].

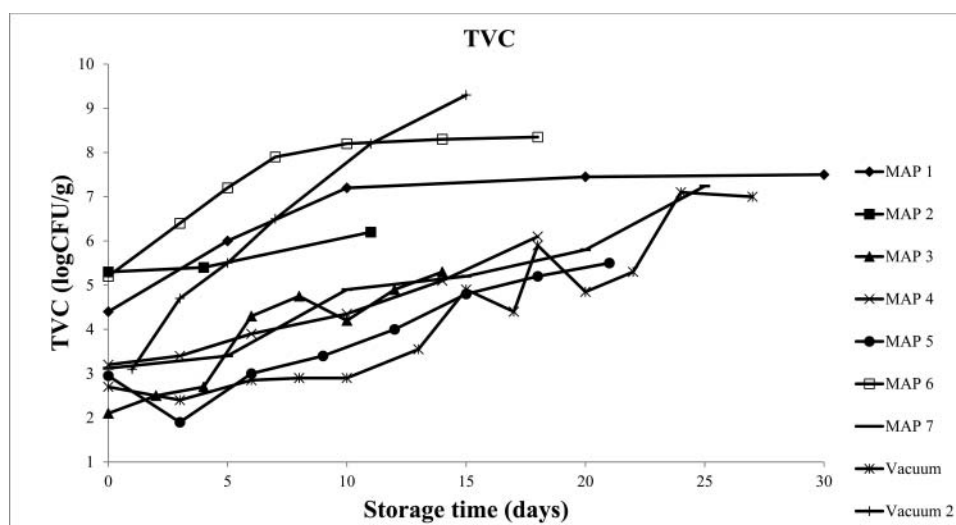


Figure 4. APC population on trout samples stored under various MA conditions [MAP 1(10% CO₂/90% N₂), Shin et al. (2008), MAP 2 (40% CO₂/60% N₂), Randell et al. (1997), MAP 3 (100% CO₂), Arahisar et al. (2004), MAP 4 (30% O₂/40% CO₂/30% N₂) on samples being slaughtered by percussive stunning, Ozogul and Ozogul, (2004), MAP 5 (5% O₂/45% CO₂/50% N₂) with the addition of oregano essential oil (0.4% v/w), Pyrgotou et al. (2010), MAP 6 (2.5% O₂/90% CO₂/7.5% N₂) on trout samples inoculated with *L. monocytogenes*, Yilmaz et al. (2009), MAP 7 (50% CO₂/50% N₂) of trout samples dry-salted, Oguzhan and Angis, (2012), Vacuum packaging of sugar-salted trout fillets, Lyhs et al. (2001), Vacuum 2 of samples fed with ketocarotenoids, Gobantes et al. (1998)].

levels were significantly lower on MAP samples (26.7 and 38.41 mg/100 g) on the 11th day of storage (Jezek and Buchtova, 2010).

A shelf life extension of three and five days was achieved with MAP 1 (30% CO₂/70% N₂) and 2 (80% O₂/20% CO₂), respectively, compared to control carp samples (three days) stored at 4 ± 0.5°C. On the 10th day, TVC and PVC were significantly lower on MAP samples (7.5, 7.4, and 12.5 and 6.7, 7.15, and 11.4 log CFU/g for TVC and PVC for MAP 1, MAP 2 and control samples, respectively). *Salmonella* spp. and *Listeria monocytogenes* were not found on the examined samples (Hudecova et al., 2010).

Fish products

Challenge studies were performed on raw and cooked surimi nuggets and stored under MAP (100% CO₂ with and

without an oxygen absorber) at 4 and 12°C. Samples remained sensorially acceptable except for samples at 12°C that were rejected on the 28th day. *L. monocytogenes* was effectively inhibited on samples under MAP with or without oxygen absorber (<6 log CFU/g on the 28th day) while on all other samples reached 7 log CFU/g at the end of storage period (Lyver et al., 1998).

The possibility to increase the shelf life of blue fish burgers with the combination of thymol (110 ppm), lemon extract (120 ppm) and grapefruit seed extract (100 ppm) and MAP (30% O₂/40% CO₂/30% N₂ for MAP 1, 50% O₂/50% CO₂ for MAP 2 and 5% O₂/95% CO₂ for MAP 3) storage at 4°C was assessed by Del Nobile et al. (2009). Combination of MAP 3 with essential oils gave the best microbiological results (4.91 and 4.95 log CFU/g for H₂S producing bacteria and psychrotolerant bacteria,

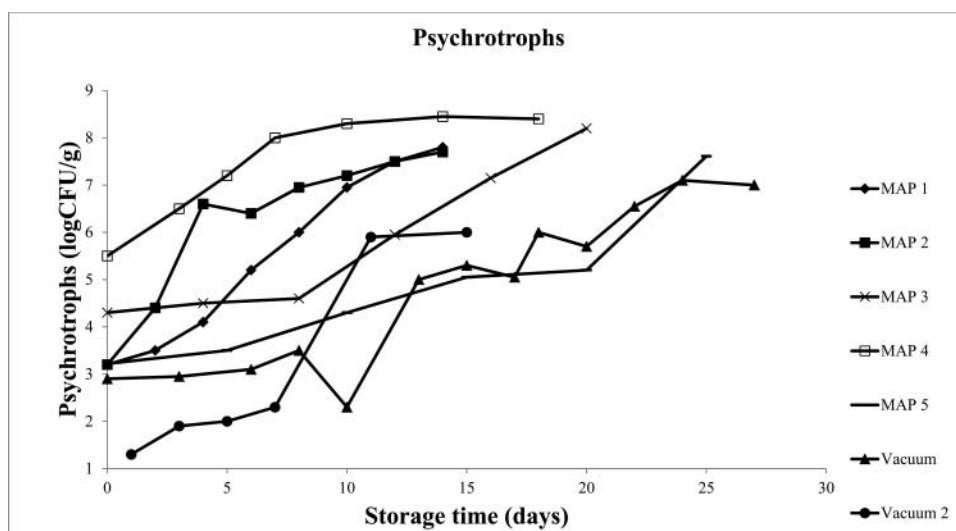


Figure 5. Psychrotroph counts on trout stored under MAP [MAP 1 (100% CO₂), MAP 2 (2.5% O₂/90% CO₂/7.5% N₂), Arahisar et al. (2004), MAP 3 (10% O₂/50% CO₂/40% N₂), Gimenez et al. (2002), MAP 4 (2.5% O₂/90% CO₂/7.5% N₂) on trout samples inoculated with *L. monocytogenes*, Yilmaz et al. (2009), MAP 5 (50% CO₂/50% N₂) of trout samples dry-salted, Oguzhan and Angis, (2012), Vacuum packaging of sugar-salted trout fillets, Lyhs et al. (2001), Vacuum 2 of samples fed with ketocarotenoids, Gobantes et al. (1998)].

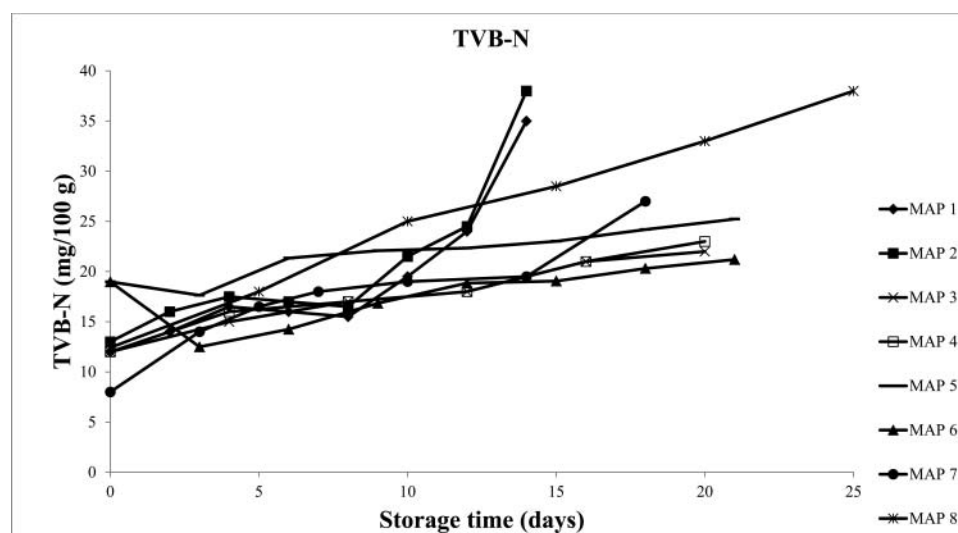


Figure 6. TVB-N content on trout samples stored in environments with modified atmosphere [MAP 1 (100% CO₂), MAP 2 (2.5% O₂/90% CO₂/7.5% N₂), Arashisar et al. (2004), MAP 3 (10% O₂/50% CO₂/40% N₂), MAP 4 (20% O₂/50% CO₂/30% N₂), Gimenez et al. (2002), MAP 5 (5% O₂/45% CO₂/50% N₂) with the addition of oregano essential oil (0.2% v/w), MAP 6 (5% O₂/45% CO₂/50% N₂) with the addition of oregano essential oil (0.4% v/w), Pyrgotou et al. (2010), MAP 7 (2.5% O₂/90% CO₂/7.5% N₂) on trout samples inoculated with *L. monocytogenes*, Yilmaz et al. (2009), MAP 8 (50% CO₂/50% N₂) of trout samples dry-salted, Oguzhan and Angis, (2012)].

respectively, on the 28th day) having the longest shelf life of 23 days but limited by unacceptable odors.

Precooked products with cod and shrimp tail origin underwent lauric acid (0.75 g/kg) pretreatment and then stored under MAP (50% CO₂/50% N₂) for 30 days at 7 ± 1°C. A synergistic effect of MAP and lauric acid on cod products was evident with APC being 4.65 log CFU/g on the 40th day, while pathogens were not detected at levels hazardous for human health. Sensory attributes of both products remained at acceptable levels for the whole 30-day storage period (Pastoriza et al., 2002).

Both atmosphere modifications (5% O₂/35% CO₂/60% N₂ for MAP 1, 30% CO₂/70% N₂ for MAP 2) preserved fish salad samples (rainbow trout, carrots, potatoes and peas) until the 14th day of storage at 4°C. TVB-N values were under the rejection limit of 25 mg/100 g (20.1 and 22.3 mg/100 g for MAP 1 and 2, respectively) while microbial populations of MAP samples were lower compared to control (8.6, 7.5, and 7.5, 8.1, 7.3, and 5.6 for APC and yeasts and molds of control, MAP 1 and 2 samples, respectively, on the 14th day) (Metin et al., 2002).

Three months shelf life extension of marinated fish salad (boiled squid, surimi, mussels, shrimp and octopus) was reached under MAP (70% CO₂/30% N₂ for MAP 1 and 50% CO₂/50% N₂ for MAP 2) storage at 2 ± 2°C. Up to the seventh month of storage levels of TBA and TVB-N for MAP samples remained below the rejection limit for the entire storage period (7.54 mg malondialdehyde/kg and 18.52 mg/100 g for TBA and TVB-N, respectively). The microbial load on MAP samples was very low for the entire storage period (Gunsen et al., 2010).

Conclusions

In most of the above described experiments, there were many parameters supporting the use of MAP at a large scale in the aquacultured fish industry. In sea bass, the

addition of thyme oil in conjunction with MAP led to a 12-day shelf life extension regarding the APC limit while high carbon dioxide atmosphere can lead to shelf life greater than 20 days. The shelf life of mussels can be prolonged by 6 to 7 days under MAP because of microbial inhibition and limitation of TMA and TVB-N production. In the case of wolf-fish a high CO₂ atmosphere led to five to seven-day shelf life extension. Various MAP applications led to chemical and microbial improvement on trout samples because of inhibition of pathogens like inoculated *L. monocytogenes*. The application of low storage temperatures (2°C and lower) and atmosphere modification without oxygen and carbon dioxide at high levels were the most effective treatments for salmon storage. High CO₂ conditions favored smoked catfish preservation. Carp had a 3 to 5 shelf life extension under MA packages compared to control.

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