IOP Conf. Series: Earth and Environmental Science **85** (2017) 012036 doi:10.1088/1755-1315/85/1/012036

Effect of modified atmosphere and vacuum packaging on TVB-N production of rainbow trout (*Oncorhynchus mykiss*) and carp (*Cyprinus carpio*) cuts

J Babić Milijašević, M Milijašević, J Đinović-Stojanović and D Vranić

Institute of Meat Hygiene and Technology, Kaćanskog 13, Belgrade, Serbia

E-mail: jelena.babic@inmes.rs

Abstract. The aim of our research was to examine the influence of packaging in modified atmosphere and vacuum on the total volatile basic nitrogen (TVB-N) content in muscle of rainbow trout (*Oncorhynchus mykiss*) and common carp (*Cyprinus carpio*), as well as to determine the most suitable gas mixtures for packing of these freshwater species. Three sample groups of trout and carp cuts were investigated. The two groups were packaged in modified atmosphere with different gas ratios: 90%CO_2+10%N_2 (MAP 1) and 60%CO_2+40%N_2 (MAP 2), whereas the third group of fish cuts were vacuum packaged. During trials, the trout and carp cuts were stored in refrigerator at 3°C±0.5°C. Determination of TVB-N was performed on 1, 4, 7, 9, 12 and 14 days of storage. The obtained results indicate that the investigated mixtures of gases and vacuum had a significant influence on the values of TVB-N in trout and carp cuts. The lowest increase in TVB-N was established in trout and carp cuts packaged in MAP 1, whereas the highest increase was established in vacuum packaged cuts. Based on the obtained results, it can be concluded that the gas mixture consisting of 90% CO_2 and 10% N_2 was the most suitable for packaging of fresh trout and carp cuts in terms of TVB-N value.

1. Introduction

Fish are highly perishable and prone to vast variations in quality due to differences in species, environmental habitats, feeding habits and action of autolysis enzymes as well as hydrolytic enzymes of microorganisms on the fish muscle. Deterioration of fish mainly occurs as a result of bacteriological activity, leading to loss of quality and subsequent spoilage [1].

The shelf life of fresh chilled fish can be extended by vacuum packaging or modified atmosphere packaging (MAP). The most frequently used packaging gases include oxygen (O_2), carbon dioxide (CO_2), nitrogen (N_2) and their combinations in different ratios. Gas mixtures with higher levels of CO_2 and N_2 as inert gases have attracted the most attention from researchers. The most frequently used CO_2 concentrations in fish packaging fall between 40 and 60%. CO_2 acts as an antimicrobial agent. It inhibits the growth of spoilage organisms, particularly *Shewanella putrefaciens*, *Pseudomonas*, *Vibrio* and *Aeromonas spp*. that produce H_2S [2]. The bacteriostatic effect of CO_2 depends on its concentration and storage temperature, and the mechanisms of action are based on changing the permeability of the bacterial membrane, inhibition of bacterial enzymes, alteration in the physico-chemical properties of the protein, and changes in the intra-cell pH. N_2 is used for displacing O_2 from packaging, decreasing oxidative rancidness and inhibiting the growth of aerobic microorganisms [3].

Content from this work may be used under the terms of the Creative Commons Attribution 3.0 licence. Any further distribution of this work must maintain attribution to the author(s) and the title of the work, journal citation and DOI.
Published under licence by IOP Publishing Ltd

The combined total amount of ammonia (NH_3), dimethylamine (DMA) and trimethylamine (TMA) in fish is called the Total Volatile Base Nitrogen (TVB-N); its content in the fish meat is commonly used as parameter for spoilage estimation, and as index for freshness of fish. It is produced during degradation of protein and non-protein nitrogen components, caused mainly by metabolic activity of fish spoilage bacteria and endogenous enzymes action [4]. NH_3 is formed by the bacterial degradation/deamination of proteins, peptides and amino-acids. It is also produced in the autolytic breakdown of adenosine monophosphate. The presence of DMA and TMA in spoiling fish is due to the bacterial reduction of TMA oxide (TMAO) which is naturally present in the living tissue of all marine and many freshwater fish species [5]. In anaerobic conditions, spoilage bacteria are able to utilize TMAO as the terminal electron acceptor which results in off-odours and -flavours due to formation of TMA. TMA and TVB-N are considered responsible for unpleasant 'fishy' odour of spoiled fish [6].

The aim of this research was to examine the influence modified atmosphere and vacuum packaging on changes in TVB-N values of rainbow trout (*Oncorhynchus mykiss*) meat and carp (*Cyprinus carpio*) meat and to determine the most suitable gas mixtures for the packaging of these two freshwater fish species.

2. Materials and Methods

2.1. Sample collection

Rainbow trout (*Oncorhynchus mykiss*) used in the study were all farmed in the same conditions and came from a trout pool located on the slopes of Zlatibor Mountain. Marketable carp (*Cyprinus carpio*) originated from a fish farm pond located in the lowland region of Serbia, where semi-intensive farming was used. In this study, two-year-old carp of average body weight of 2.5 kg were used. Three sample groups of cleaned trout and carp cuts were formed. Group 1 cuts were vacuum packaged and were used as the control. The other two sample groups were packaged in modified atmospheres with different gas ratios: MAP 1: 90% CO_2+10% N_2 and MAP 2: 60% CO_2+40% N_2. The machine used for packaging was Variovac (Variovac Primus, Zarrentin, Germany), and the material used for packaging was foil OPA/EVOH/PE (oriented polyamide/ethylene vinyl alcohol/polyethylene, Dynopack, Polimoon, Kristiansand, Norway) with low gas permeability (degree of permeability for O_2 - 3.2 cm^3/m^2/day at 23°C, for N_2 - 1 cm^3/m^2/day at 23°C, for CO_2 - 14 cm^3/m^2/day at 23°C and for steam 15 g/m^2/day at 38°C). The ratio of gas:fish cuts in the packages was 2:1. All fish cuts were stored in the same conditions at 3°C±0.5°C and on 1, 4, 7, 9, 12 and 14 days of storage, chemical testing was performed.

2.2. Chemical analysis

The total volatile basic nitrogen (TVB-N) was determined by using the official steam distillation method according to Commission Regulation (EC) 2074/2005 and was expressed as mg TVB-N/100 g.

2.3. Statistical analysis

The mean values and standard deviations were calculated by using column statistics with the processing of six values for each analysed group. Significant differences between groups were calculated by using one-way ANOVA analysis by Tukey's comparative test in the program Microsoft Office Excel (2010). Differences were considered as significant when p value was < 0.05.

3. Results and Discussion

At the beginning of the study, concentrations of TVB-N in carp steaks as well as in trout cuts were practically identical ($P > 0.05$). During the storage period, TVB-N value increases were observed in all experimental groups. As shown in figures 1 and 2, TVB-N values were strongly affected by the packaging atmosphere used. Increases in TVB-N values of carp and trout cuts in the different packaging atmospheres followed the order: MAP 1 < MAP 2 < vacuum.

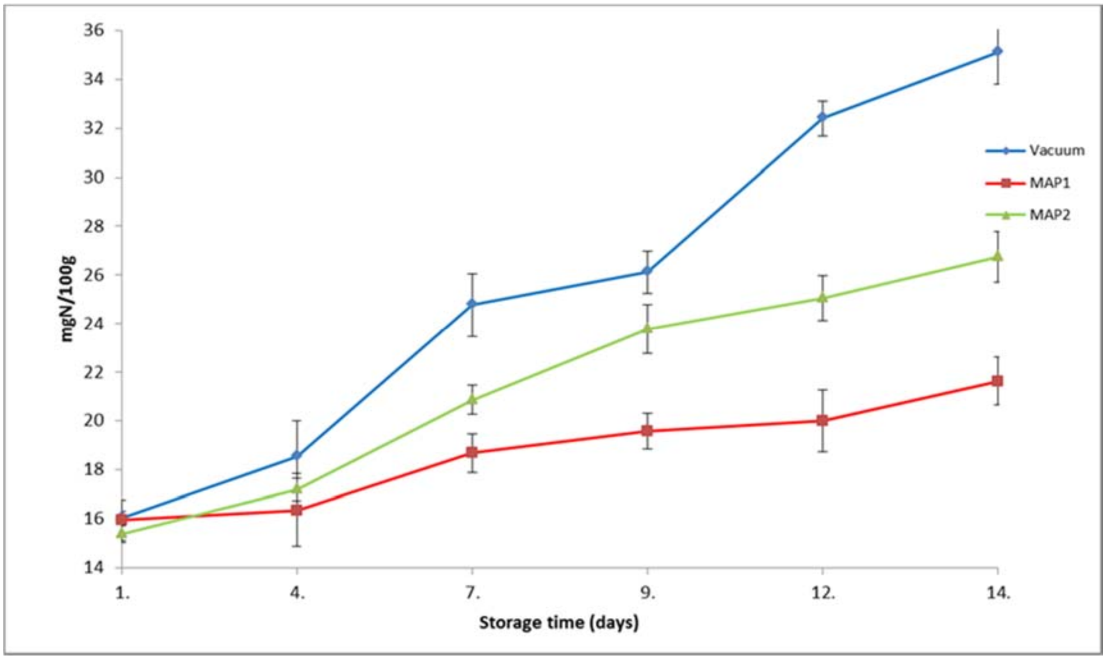

Figure 1. TVB-N value of common carp steaks packaged under different conditions during the storage period.

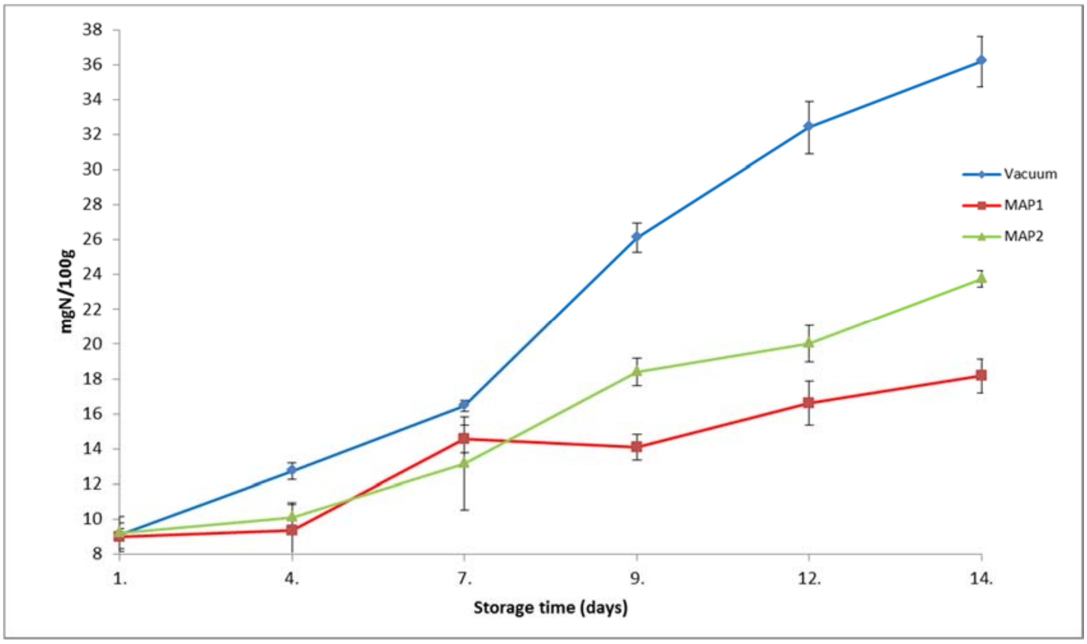

Figure 2. TVB-N value of trout cuts packaged under different conditions during the storage period.

TVB-N levels in fish packaged under MAP 1 changed to lesser extent comparing to those levels in fish packaged under MAP 2 and vacuum. It is interesting to note that up to 7 days of storage, there were no statistically significant differences ($p > 0.05$) in TVBN values between fish packaged in MAP

1 and MAP 2. This may be attributed to the fact that when the total bacteria count of fish flesh is still relatively low, a gas mixture of 60% CO_2 + 40% N_2 is adequate to inhibit the microbial activity, as exhibited by low TVBN values. When microbial populations increase, a higher concentration of CO_2 is required to inhibit the microbial activity and the subsequent spoilage.

According to Masniyom *et al.* [7], a high CO_2 concentration potentially inhibited the growth of mainly Gram negative microorganisms and decreased the deamination capacity of bacteria, resulting in lower production of volatile compounds. The same observations were reported by Milijašević *et al.* [8] and Babić *et al.* [9] for carp and trout stored under MAP, which support the results of the present study.

The TVB-N limit from 25 to 35 mg N/100g has been recommended by some researchers as an indicator for rejecting commercial fresh whole fish and processed fish products [4]. However, no limit for acceptability of TVB-N in common carp and rainbow trout has been established by the Commission Regulation (EC) 2074/2005. Examining the effect of several different gas mixtures on the shelf life of rainbow trout fillets, Gimenez *et al.* [10] found that MAP is very effective in preventing the production of TVB-N. These authors recommended a value of 25 mg N/100g as the highest acceptability level of TVB-N for rainbow trout [10]. In our research, this limit was exceeded in control fish cuts on day 9, while TVB-N values in MAP 1 and MAP remained below this recommended limit. In their research, Ježek and Buhtova [11] recommend 20 mg N/100g in carp meat as the highest acceptable limit for TVB-N. In our current study, this limit was exceeded in control fish cuts (day 7), MAP2 fish cuts (day 9), as well as in MAP1 fish cuts (day 14).

4. Conclusion

Based on the obtained results, it can be concluded that the gas mixture consisting of 90% CO_2 and 10% N_2 was the most suitable for packaging of fresh trout and carp cuts in terms of TVB-N value.

References

[1] Chytiri S, Chouliara I, Savvaidis I N and Kontominas M G 2004 *Food Microbiol.* **21** 157
[2] Debevere J and Boskou G 1996 *Int J. Food Microbiol.* **31** 221
[3] Farber J M 1991 *J. Food Prot.* **54** 58-70
[4] Connell J J 1990 *Control of fish quality* (Oxford: Fishing News Books) p 122
[5] Huss H H 1995 FAO Fisheries technical paper – 348 (Available at: http://www.fao.org/docrep/V7180E/V7180E00.HTM)
[6] Gram L and Huss H H 1996 *Int J. Food Microbiol* **33** 121
[7] Masniyom P Benjakul S and Visessanguan W 2002 *J. Sci. Food Agric.* **82** 873
[8] Milijašević M, Babić J, Baltić MŽ, Spirić A, Velebit B, Borović B and Spirić D 2010 *Tehnologija mesa* **51**(1) 66
[9] Babić J, Dimitrijević M, Milijašević M, Đorđević V, Petronijević R, Grbić S and Spirić A 2014 *Hem. Ind.* **68** 69
[10] Gimenez B P, Roncales P and Beltran J A 2002 *J. Sci Food Agric.* **82** 1154
[11] Ježek F and Buchtová H 2010 *Acta Vet. Brno* **79** 117