



The influence of modified atmosphere and vacuum packaging on selected chemical freshness parameters of perch (*Stizostedion lucioperca*) and silver carp (*Hypophthalmichthys molitrix*) fillets

Milan Milijašević^{1*} , Jelena Babić Milijašević¹ , Slobodan Lilić¹ , Vesna Dorđević¹ , Branka Borović¹ , Aleksandra Nikolić¹  and Slavica Vesković Moračanić¹ 

¹ Institute of Meat Hygiene and Technology, Kačanskog 13, 11000 Belgrade

ARTICLE INFO

Keywords:

Fish
Modified atmosphere packaging
Vacuum
Perch
Silver carp

ABSTRACT

The purpose of packing food in modified atmospheric conditions is to extend its sustainability by preventing both biochemical processes and growth of spoilage bacteria. 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 perch (*Stizostedion lucioperca*) and silver carp (*Hypophthalmichthys molitrix*). Three groups of perch and silver carp fillets were investigated. The first two groups were packaged in modified atmosphere with different gas ratios: 70%CO₂+30%N₂ (group I) and 50%CO₂+50%N₂ (group II), whereas the third, control group, (group III) was vacuum packaged. During the trial, the packaged fish was stored at 3°C. Determination of TVB-N was performed on days 1, 4, 7, 9 and 14 of storage. The lowest increase in TVB-N was established in perch and silver carp fillets from group I, whereas the highest increase was established in group III. Statistically significant differences ($p < 0,001$) between the mean TVB-N in perch (group I: 17.00±0.94; group II: 25.00±0.91 and group III: 36.18±2.65 mg N/100 g) and silver carp fillets (group I: 20.64±1.45; group II: 26.74±0.31 and group III: 34.10±1.75 mg N/100 g) was established on day 14. Based on the obtained results, it can be concluded that, in terms of TVB-N content, the gas mixture consisting of 70% CO₂ and 30% N₂ was the most suitable of the two gas mixtures for packaging fresh perch and silver carp fillets.

1. Introduction

The fact that fresh fish is a highly perishable food item (pH > 6.0; water activity > 0.98) has influenced producers to focus on finding the optimal method of fish preservation. However, in recent years, consumers around the world have increasingly demanded that fresh fish be available at all times,

as it has the most desirable sensory characteristics (Babić Milijašević et al., 2023; Esteves et al., 2021). This trend has led to the development of an efficient modified atmosphere packaging (MAP) concept, which ensures a longer shelf life and preserves key freshness parameters (Gimenez et al., 2002).

Gas mixtures with high concentrations of CO₂ and N₂ have attracted the most attention from

*Corresponding author: Milan Milijašević, milan.milijasevic@inmes.rs

Paper received Jun 26th 2025. Paper accepted Jun 25th 2025.

The paper was presented at the 63rd International Meat Industry Conference “Food for Thought: Innovations in Food and Nutrition” – Zlatibor, October 05th–08th 2025.

Published by Institute of Meat Hygiene and Technology – Belgrade, Serbia.

This is an open access article CC BY licence (<http://creativecommons.org/licenses/by/4.0>)

researchers dealing with fish packaging issues over the past decade (Zhang *et al.*, 2021). However, the impact of modified atmosphere packaging on the shelf life of fresh fish and the most suitable gas mixture depends on the type of fish being packaged, fat content, initial microbial contamination, handling after catch, the volume ratio of gas to product in the package, and most importantly, the packaging method and storage conditions (Sivertsvik *et al.*, 2002; Bøknæs *et al.*, 2000). In some cases, MAP can negatively affect the quality of packaged fish due to the dissolution of CO₂ in the fish meat, leading to the formation of carbonic acid (Babić Milijašević *et al.*, 2018). At lower pH values, the water-binding capacity of the fish meat decreases, causing the release of meat juice in the package, which serves as an ideal medium for the growth of spoilage microorganisms (Sivertsvik *et al.*, 2002; Goulas and Kontominas, 2007). For these reasons, it is essential to determine the optimal gas ratio in the mixture based on the characteristics of the packaged product and the packaging system.

The level of total volatile basic nitrogen (TVB-N) is considered a chemical indicator of fish freshness. TVB-N consists of compounds responsible for the unpleasant odour and taste of fish meat, including ammonia, dimethylamine (DMA), trimethylamine (TMA), amines formed through the decarboxylation of amino acids, as well as other nitrogen compounds that become volatile in their alkaline form (Debevere and Boskou, 1996; Ruiz-Capillas and Moral, 2005). Ammonia is produced by bacterial deamination of proteins, peptides, and amino acids, as well as the autolytic degradation of adenosine monophosphate (AMP). Dimethylamine and trimethylamine result from the breakdown of trimethylamine oxide (TMAO), a compound that plays a significant role in osmoregulation and whose presence has been confirmed in all marine and many freshwater fish species. Due to the activity of endogenous fish enzymes, TMAO is degraded, leading to the formation of DMA and formaldehyde (Huss, 1995).

The aim of this study was to examine the impact of modified atmosphere packaging and vacuum packaging on changes in the TVB-N in the meat of perch (*Stizostedion lucioperca*) and common silver carp (*Hypophthalmichthys molitrix*), and to determine the most suitable gas mixtures for packaging these two freshwater fish species.

2. Materials and methods

The perch (*Stizostedion lucioperca*) used in the experiment were raised under identical conditions and originated from a semi-intensive farming pond. For the study, 40 one-year-old perch with a mean weight of 320 g were selected. The fish were transported alive from the farm to the processing facility, where they were placed in a reception tank and then stunned using electric current. Slaughtering and evisceration were performed using an automatic device, while carcasses were washed manually under running water.

The silver carp (*Hypophthalmichthys molitrix*) came from a fish farm where a semi-intensive farming method was applied. For the study, 10 two-year-old silver carp with a mean weight of 2.5 kg were used. The silver carp were transported alive to the processing facility, where they were stunned, slaughtered, and descaled.

Fish carcasses were then cut into 4 cm thick fillets, with each carcass yielding 6 fillets. Three sample groups were formed, each consisting of 18 cleaned perch or silver carp fillets. The first two groups were packed using MAP with different gas ratios: 70% CO₂ + 30% N₂ (Group I) and 50% CO₂ + 50% N₂ (Group II), while the third, control group was vacuum-packed. The samples were packaged using a Variovac machine (Variovac Primus, Zarrentin, Germany). The packaging material was OPA/EVOH/PE film (oriented polyamide/ethylene vinyl alcohol/polyethylene, Dynopack, Polimoon, Kristiansand, Norway) with low gas permeability (O₂ permeability: 3.2 cm³/m²/day at 23 °C; N₂: 1 cm³/m²/day at 23 °C; CO₂: 14 cm³/m²/day at 23 °C; and water vapor: 15 cm³/m²/day at 38 °C). The gas-to-sample ratio in the packaging was 2:1. Throughout the study, all packages of fish were stored under identical conditions at 3 °C. Determination of TVB-N was performed on days 1, 4, 7, 9 and 14 of storage.

The total volatile basic nitrogen (TVB-N) was determined using the EU reference method, as specified in *Commission Regulation (EC)* (2005).

For statistical analysis (mean value, variation measures, variance analysis, and t-test), the Microsoft Office Excel 2016 software package was used.

3. Results and discussion

The mean TVB-N levels in perch packed in the modified atmospheres and in vacuum packaging are presented in Figure 1. Based on the displayed

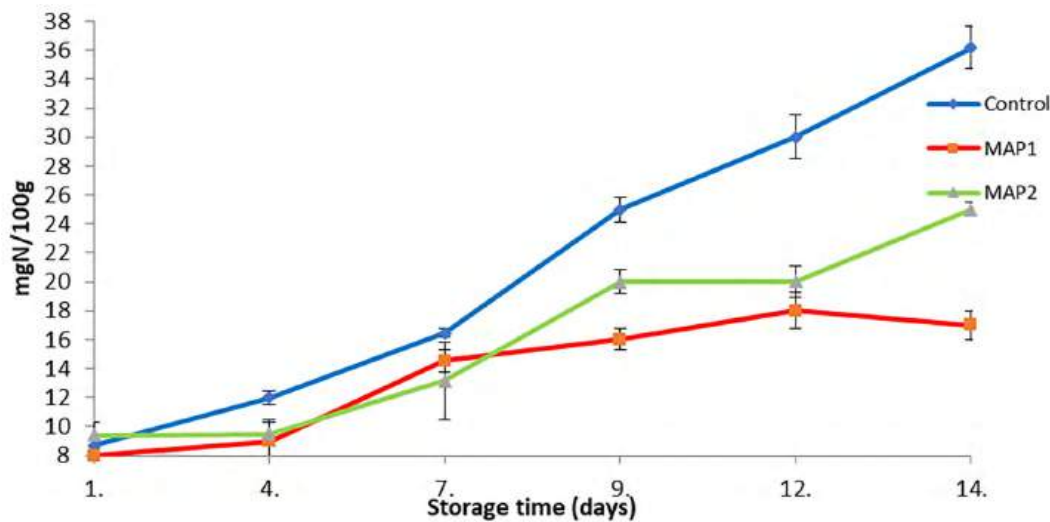


Figure 1. Mean TVB-N levels in perch packaged in modified atmosphere (MAP) and vacuum (control).

results, it can be observed that on day 1, the mean TVB-N content in all three groups of perch (8.00 ± 0.32 , 9.40 ± 0.46 , and 8.70 ± 0.31 mg N/100 g) did not differ significantly ($p > 0.05$). The mean TVB-N content in group I (14.55 ± 0.60 mg N/100 g) and group II (13.15 ± 0.98 mg N/100 g) after seven days of storage showed a statistically significant difference ($p < 0.001$) compared to the mean TVB-N content in group III (16.46 ± 1.00 mg N/100 g). No statistically significant difference ($p > 0.05$) was found between TVB-N levels in groups I and II on day 7. On day 14, a statistically significant difference ($p < 0.001$) was established between the mean TVB-N levels in group I (17.00 ± 0.93 mg N/100 g), group II (25.00 ± 0.81 mg N/100 g), and group III (36.18 ± 2.65 mg N/100 g).

The mean TVB-N levels in silver carp fillets packed in the modified atmosphere and vacuum packaging are presented in Figure 2.

On day 1, no statistically significant difference ($p > 0.05$) was found between the mean TVB-N (16.15 ± 0.84 , 15.45 ± 0.78 , and 15.05 ± 0.27 mg N/100 g) in silver carp fillets. On day 7, the mean TVB-N value in group I (19.01 ± 0.78 mg N/100 g) was the lowest and showed a statistically significant difference ($p < 0.05$) compared to group III (24.90 ± 1.74 mg N/100 g). The difference between TVB-N levels in group II (20.86 ± 0.75 mg N/100 g) compared to group III was also statistically significant at $p < 0.001$. No statistically significant difference ($p > 0.05$) was found between the mean TVB-N levels in groups I and II on day 7. On day 14, the lowest mean

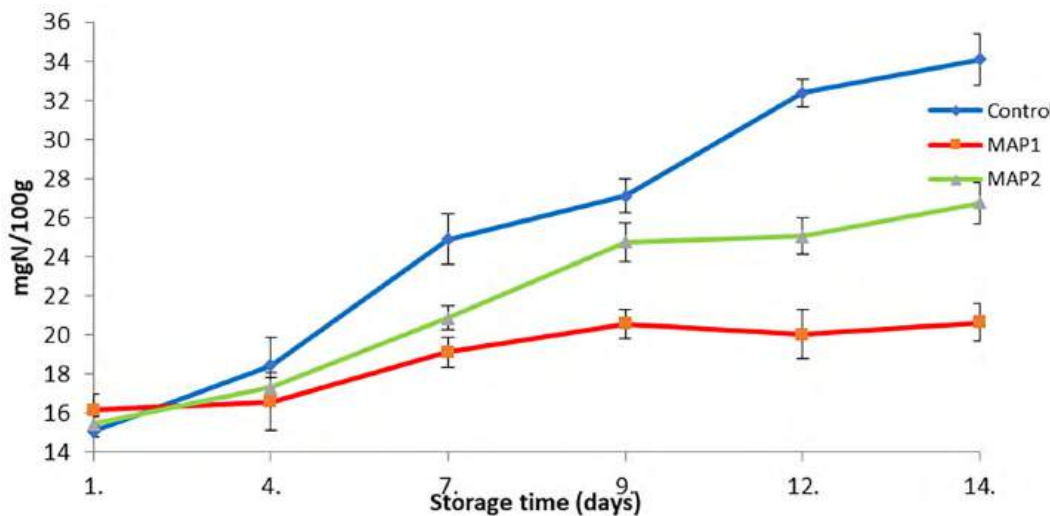


Figure 2. Mean TVB-N levels in silver carp packaged in modified atmosphere (MAP) and vacuum (control).

TVB-N level was found in silver carp from group I (20.64 ± 1.48 mg N/100 g), whereas the highest was observed in group III (34.10 ± 1.75 mg N/100 g). The mean TVB-N level in group II on day 14 was 26.74 ± 0.31 mg N/100 g. The differences between the mean TVB-N levels in all three groups of silver carp fillets on day 14 were statistically significant ($p < 0.001$).

Arahisar *et al.* (2004) found that the initial TVB-N level in trout fillets was 12 mg N/100 g, while Ježek and Buhtova (2010) reported an initial TVB-N of 16.25 ± 0.79 mg N/100 g in carp fillets. The differences in TVB-N levels in the fish compared to those measured in our study could be attributed to variations in the non-protein nitrogen content of fish meat, which depends on diet, time of catch, fish size, and microbiological quality (Connell, 1990).

The observed differences in TVB-N levels between our MAP test groups can be explained by the higher percentage of CO₂ in the gas mixture used to pack group I. The primary role of CO₂ in MAP packaging technology is to inhibit microbial growth, especially spoilage bacteria, which produce volatile nitrogen compounds through their metabolic activity.

There is no established limit for TVB-N in perch meat in the literature, but we consider 25 mg N/100 g as a critical threshold. In our study, TVB-N levels in MAP-packed perch (groups I and II)

remained below this threshold throughout the storage period, whereas in vacuum-packed perch (group III), the mean TVB-N reached 36.18 ± 2.65 mg N/100 g on day 14, significantly exceeding the recommended limit for perch freshness.

Milijašević *et al.* (2010) examined the inhibitory effect of CO₂ on TVB-N content in carp (*Cyprinus carpio*) fillets, confirming that the lowest TVB-N level was recorded in carp fillets packed in a modified atmosphere containing 100% CO₂. Similarly, Ježek and Buhtova (2010) found lower TVB-N in carp (*Cyprinus carpio*) fillets packed in MAP (69% N₂ + 25% CO₂ + 5% O₂ + 1% CO) compared to fillets stored under refrigeration at 2 °C. These authors recommend a maximum acceptable TVB-N limit of 20 mg N/100 g in carp meat.

In our study, TVB-N values in MAP-packed silver carp fillets (groups I and II) reached this recommended limit on day 14, while in vacuum-packed fish (group III), the limit was reached on day 7.

4. Conclusion

Based on the obtained results, it can be concluded that, in terms of the selected chemical parameter, TVB-N, the most suitable gas mixture for packaging fresh perch and silver carp fillets was the mixture containing 70% CO₂ and 30% N₂.

Disclosure Statement: No potential conflict of interest was reported by the authors.

Funding: The research results presented in this paper were funded by the Ministry of Science, Technological Development, and Innovation of the Republic of Serbia, based on the Agreement on the Implementation and Financing of Scientific Research Work of the Scientific Research Organization (NIO) in 2025, No. 451-03-136/2025-03/200050, dated February 4, 2025.

References

- Arashisar, Š., Hisar, O., Kaya, M. & Yanik, T. (2004). Effect of modified atmosphere and vacuum packaging on microbiological and chemical properties of rainbow trout (*Oncorhynchus mykiss*) fillets. *International Journal of Food Microbiology*, 97, 209–214.
- Babic Milijasevic, J., Milijasevic, M., Lilic, S., Djinic-Stojanovic, J., Nastasijevic, I., & Geric, T. (2023). Effect of vacuum and modified atmosphere packaging on the shelf life and quality of gutted rainbow trout (*Oncorhynchus mykiss*) during refrigerated storage. *Foods*, 12(16), 3015.
- Babić Milijašević, J., Milijašević, M., Vranić, D., Đinović-Stojanović, J., Lilić, S. & Korićanac, V. (2018). Effect of modified atmosphere packaging on the shelf life of chilled Common carp (*Cyprinus carpio*) steaks: Chemical and sensory attributes. *Czech Journal of Food Sciences*, 36, 221–226.
- Bøknæs, N., Østerberg, C., Nielsen, J. & Dalgaard, P. (2000). Influence of freshness and frozen storage temperature on quality of thawed cod fillets stored in modified atmosphere packaging. *Lebensmittel-Wissenschaft & Technologie*, 33, 244–248.
- Commission Regulation (EC) (2005). Commission regulation (EC) 2074/2005 of 5 December 2005 laying down implementing measures for certain products under Regulation (EC) No 853/2004 of the European Parliament and of the Council and for the organisation of official controls under Regulation (EC) No 854/2004 of the European Parliament and of the Council and Regulation (EC) No 882/2004 of the European Parliament and of the Council, derogating from Regulation (EC) No 852/2004 of the European Parliament and of the Council and amending Regulations (EC) No 853/2004 and (EC) No 854/2004.

- Connell, J. J. (1990). Methods of assessing and selecting for quality. In: Connell J.J. (ed.) Control of fish quality. *Fishing News Books*, Oxford, 1990, pp. 122–150.
- Devere, J. & Boskou, G. (1996). Effect of modified atmosphere packaging on the TVB/TMA producing microflora of cod filets. *International Journal of Food Microbiology*, 31, 221–229.
- Esteves, E., Guerra, L. & Anibal, J. (2021). Effects of vacuum and modified atmosphere packaging on the quality and shelf-life of gray triggerfish (*Balistes capriscus*) filets. *Foods*, 10, 250.
- Gimenez, B., Roncales, P. & Beltran, J. A. (2002). Modified atmosphere packaging of filleted rainbow trout. *Journal of the Science of Food and Agriculture*, 82, 1154–1159.
- Goulas, A. E. & Kontominas, M. G. (2007). Effect of modified atmosphere packaging and vacuum packaging on the shelf-life of refrigerated chub mackerel (*Scomber japonicus*): biochemical and sensory attributes. *European Food Research and Technology*, 224, 545–553.
- Huss, H. H. (1995). Quality and quality changes in fresh fish. *FAO Fisheries Technical Paper* 348, FAO.
- Ježek, F. & Buchtová, H. (2010). Shelf-life of chilled muscle of common carp (*Cyprinus carpio* L.) packaged in carbon monoxide enriched modified atmosphere. *Acta Veterinaria Brno*, 79, 117–125.
- Milijašević, M., Babić, J., Baltić, M.Ž., Spirić, A., Velebit, B., Borović, B. & Spirić, D. (2010). Uticaj različitih smeša gasova na promene nekih mikrobioloških i hemijskih parametara u odrescima šarana (*Cyprinus carpio*) upakovanih u modifikovanu atmosferu. *Tehnologija mesa* 51(1), 66–70.
- Milijašević, M., Đorđević, V., Nikolić, A., Janković, V., & Babić Milijašević, J. (2024). Effect of vacuum packaging on microbial and sensory quality indicators of cold-smoked freshwater fish. *Meat Technology*, 65(2), 87–92.
- Popov, S. (2007). Biotehnologija-izazov za prehrambenu industriju. *Hemijska Industrija*, 61(5), 246–250.
- Ruiz-Capillas, C. & Moral, A. (2005). Sensory and biochemical aspects of quality of whole bigeye tuna (*Thunnus obesus*) during bulk storage in controlled atmospheres. *Food Chemistry*, 89(3), 347–354.
- Sivertsvik, M., Jeksrud, W. K., & Rosnes, J. T. (2002). A review of modified atmosphere packaging of fish and fishery products – significance of microbial growth, activities and safety. *International Journal of Food Science & Technology*, 37, 107–127.
- Zhang, J., Li, Y., Yang, X., Liu, X., Hong, H. & Luo, Y. (2021). Effects of oregano essential oil and nisin on the shelf life of modified atmosphere packed grass carp (*Ctenopharyngodon idellus*). *Food Science and Technology*, 147, 111609.

Authors info

Milan Milijašević, <https://orcid.org/0000-0003-4269-236X>

Jelena Babić Milijašević, <https://orcid.org/0009-0003-8923-7046>

Slobodan Lilić, <https://orcid.org/0009-0004-9452-0361>

Vesna Đorđević, <https://orcid.org/0009-0008-5187-4089>

Branka Borović, <https://orcid.org/0000-0003-0371-0991>

Aleksandra Nikolić, <https://orcid.org/0009-0009-0012-3418>

Slavica Vesković Moračanin, <https://orcid.org/0000-0002-6904-9510>