

## **Combined Effects of High-Pressure Processing and Marination on The Quality of Herring (*Clupea harengus*) Fillets**

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### **Abstract**

In this study, it was focused on the combined effects of marination and high pressure processing (HPP) treatment on herring fillets. For this purpose, herring fillets were marinated with 3% acetic acid and 6% NaCl solution at 4°C for three days. After ripening process marinated fish samples were vacuum packaged and treated with HPP in different pressure levels (100, 300, and 500 MPa) and pressure holding times (5 and 10 min.). One group was left untreated as control. All samples were stored at 4±1°C for 90 days. During the storage, pH, TVB-N and TMA-N values were assessed. According to results the samples treated with 300 and 500 MPa showed lower results than the groups treated with 100 MPa and the control. Moreover, 500 MPa HPP treatment had the best effects on the maintaining the quality of marinated herring. It can be concluded that HPP treatment can be used to preserve good quality of marinated fish for long term storage.

**Keywords:** Marination, non- thermal processing, high pressure processing, fish quality

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### **INTRODUCTION**

Fish demand is increasing worldwide, mostly people in developing countries consumed fish to complete their protein needs (James, 1986). Fish is a perishable product which inclined it to quickly spoil through microbes. Some basic reasons which help quick propagation of microbes known as lipid contents, less collagen and relatively high quantity of soluble nitrogen composites in the muscle. After fish death many biochemical variations occurred in fish muscle due to activity of endogenous enzymes. Many proteases like peptidases and cathepsins used for breakdown of muscles protein during post-mortem storage of fish (Sherekar et al., 1986; Sicbert, 1958). Fish and other seafood can be preserved for long time by using different methods such as salting, drying, smoking, freezing and marination. Immediately after harvesting fish can be marinated for enhancing storage and transportation time. Marination is an oldest common method of fish preservation in Europe. “Marinade” is a term which is used for fresh, freeze whole fish or fish pieces after treated by salt, sauces, oil and placed into brines. Eatable natural acid, mostly acetic acid is also used for fish marination (Meyer, 1965).

During marination salt and Acetic acid are used to stop the activity of enzymes and microbes and also improve the taste of fish with prolong shelf life (Meyer, 1965; Mc Lay, 1972). Fish can be semi-preserved through marination which is eatable without cooking and required only little preparation before serving (Fuentes et al., 2010).

Due to semi preservation they required industrial techniques to extend shelf life for commercial storage of fish. Marination preservation method based on mixture of salt and acetic acid. Smoothness and other physical properties of marinated fish product will be enhanced by appropriate penetration of salts, acids and additional elements (Yashoda et al., 2005). Concentration of salts and acids in marination mixture depends on many aspects involving fish species, fish size, fish weight, lipid content, thickness of fillet and surrounding temperature. Acetic acid and salts used for breakdown of proteins which concluded as stiffness in fish meat and not as much of susceptible to swelling (Shenderyuk and Bykowski, 1990). In this condition fish meat capacity of water holding become less and water from fish flesh discharged into marinating mixture. Some additional components along water including lipids, proteins and minerals drawn-out into the marinating mixture which leads to less weight yield and poor quality of fish product. Techniques which can be used to extend the shelf life of marinated fish product at industrial level for storage and packaging (Ozden and Erkan, 2006; Sallam et al., 2007; Gunsen et al., 2010, 2011; Ucak et al., 2019; Ucak and Gökoğlu, 2020) involve addition of flavors and some plant extracts (Cadun et al., 2008; Sen and Temelli, 2003; Guldas and Hecer, 2012; Kucukgulmez, 2012) and pasteurization (Kilinc and Cakli, 2005).

Advanced non-thermal techniques also used for achieving consumer requirements of less treated foods, including resound technology (Guimaraes et al., 2018), cold plasma (Coutinho et al., 2018), pulsed electric field (Odriozola-Serrano et al., 2013), ohmic heating (Costa et al., 2018), supercritical carbon dioxide (Amaral et al., 2017; Amarala et al., 2018) and high pressure processing technology (HPP) (Figueiredo et al., 2015; Teixeira et al., 2014; Ucak et al., 2018)). High pressure processing is commercially used for pasteurization of food products such as meat, seafood and fruit juices (Medina-Meza et al., 2014). This technique is used to deactivate enzymes and spoiling microorganisms at less temperature with slight modifications in surface consistency, nutrients, taste and color of the product (Smelt, 1998; Thakur and Nelson, 1998; Considine et al., 2008). High-pressure processing is a non-thermal technique used to achieve consumer's requirements of less treated suitable high quality products with original taste (Oey et al., 2008; Patras et al., 2009). The basic target of HPP is bacterial cell membranes (Smelt, 1998), so it interrupts process of chemical permeability which occurred through membrane. Due to this damage cell components transformed to waste, pH change, enzymes become denatured, and eventually lead to cell death (Smelt et al., 2001). In this study it was aimed to evaluate the quality of marinated herring fillets treated with high pressure processing during cold storage.

## **MATERIAL AND METHODS**

### **Preparation of fish marinade an HPP treatment**

Herring (*Clupea harengus*) fillets were provided from fish market in Germany (Quakenbrück). Fish fillets were transported in ice boxes to the laboratory of German Institute of Food Technologies. Then fillets were stored at -20°C until using. For the marination process 3% acetic acid (v/v)+6% NaCl (w/v) solution was prepared in the glass jars. The skins of thawed fish fillets were removed aseptically and rinsed with distilled water. Fish-to-solution ratio was arranged as 1:1.5 (w/v) and fish marinades were placed into glass jars.

The ripening process was performed 4°C for 3 days. The vacuum-packed marinated fish were treated with a high-pressure test system (WAVE 6000/55HT; NC Hyperbaric, Burgos, Spain) possessing a 55-L chamber and a maximum pressure level of 600 MPa. 100, 300 and 500 MPa pressure levels were applied for 5 and 10 min. Control was left as untreated. All samples were stored at 4°C for 3 months and periodically analyzed.

### **Analyses**

The pH value of the samples was determined by dipping the pH-meter probe into the fish homogenates (1:1, w:v, fish:distilled water). For the determination of total volatile basic nitrogen (TVB-N) the method of Schormuller (1968) was used. The results were expressed as mg nitrogen/100 g sample. Trimethylamine (TMA-N) analysis was performed according to the method of Schormüller (1968).

### **Statistical analysis**

All measurements were carried out in triplicate and data were subjected to Analysis of Variance (ANOVA) and Duncan's multiple range tests using the SPSS Version 18.0 statistical package (SPSS Inc., Chicago, IL, USA). Differences were regarded statistically significant at  $P < 0.05$ .

## **RESULTS AND DISCUSSION**

### **pH**

When fish become spoil due to the activity of microorganisms, some compounds discharge which has bad smell and taste like ammonia and hydrogen sulphide. Due to excretion of these compounds pH of fish sample will be increased (Gennari et al., 1999). High pressure processing reduces existing acidic groups in muscle proteins which causes high level of pH (Angsupanich and Ledward, 1998; Ma et al., 2007).

Herring fillets samples were taken on end of the trial and pH value were observed at 0, 15, 30, 45, 60, 75 and 90 days at  $4 \pm 1^\circ\text{C}$ . The effect of storage on marinated herring fillets treated with HPP samples pH is given in Table 1. The pH value in herring fillets found increased with storage period and pH value was found highest at day 90 (4.62) as compared to pH at day 0 (4.40) ( $P < 0.05$ ). The groups B, C, D, E, F, and G had lower pH values than the control group in all days. The lowest pH value was observed in group G (4.37) followed by F (4.43), E (4.47), D (4.53), C (4.53), B (4.57) and A (4.62) after 90 days of storage. In contrast to our findings Bindu et al. (2013) performed an experiment on *Fenneropenaeus indicus*, who observed that different levels of HPP had significant results ( $P < 0.05$ ). It might be opposite to our study because their pH values increased as pressure was increased while in our study G group with highest pressure has lowest pH value. Increment of pH with increasing pressure was also observed in another study on minced albacore muscle. Escalation in pH is occurred because HPP encouraged protein breakdown through unfolding and ionization of protein (Morild, 1981; Yamamoto et al., 1994).

### **Total volatile basic nitrogen (TVB-N)**

Total volatile basic amines (TVB-N) are commonly used process to check the quality of seafood. TVB-N value in herring fillets was observed at 0, 15, 30, 45, 60, 75 and 90 days after storage at 4°C (Table 1). At 0 day, TVB-N value of herring fillets of control group was (17.60 mg N/100 g) and increased in all samples during the storage and at the end of storage on 90th day TVB-N value was found highest (56.70 mg N/100 g) in control group ( $P<0.05$ ). The control group showed the highest TVB-N value on all days. At 90th day, it was found that group G had significantly ( $P<0.05$ ) lowest TVB-N value (28.40) followed by F (31.76), C (42.94), E (43.52), D (47.91), B (51.91) and control group (56.70). It was noticed that the TVB-N value of the marinated herring fillet decreased with increasing HPP level on all days. The results obtained revealed that the quality of the marinated herring fillet treated with HPP used in the research was effective during storage time. Bindu et al., 2013 worked on *F. indicus*.

He observed that Pressure has a significant effect on TVB-N values during storage. It was observed that TVB-N values were decreased with increasing pressure level and lowest value was observed on last day of experiment at highest level of pressure which was 600 MPa (Bindu et al., 2013). In the present study we also have the same observation that group G with highest pressure level has lowest TVB-N value as compared to other groups.

### **Trimethylamine (TMA-N)**

Trimethylamine (TMA-N) is produced in spoiled fish with typical fishy smell and bitter taste. This process occurs due to some specific spoilage bacteria which have the ability to produce high quantity of spoilage causing compounds known as trimethylamine. Trimethylamine is also produced from non-protein nitrogen composite found in seafood known as trimethylamine oxide (TMAO). Trimethylamine oxide transformed into trimethylamine through bacterial action and endogenous enzymatic action (Regenstein et al., 1982). High pressure processing decreased trimethylamine through restricting protein breakdown (Hernández-Andrés et al., 2005). TMA-N value in herring fillets was observed at 0, 15, 30, 45, 60, 75 and 90 days after storage at 4°C (Table 1). At 0 day, TMA-N value of herring fillets of control group sample was (2.51 mg/100 g) and increased in all samples during the storage and at the end of storage on 90th day. TMA-N value was found highest (13.24 mg/100 g) in control group ( $P<0.05$ ). The control group showed the highest TMA-N value on all days. At 90th day, it was found that group G had the significantly ( $P<0.05$ ) lowest herring fillets TMA-N value (4.25) followed by D (5.13), F (5.29), E (5.62), C (6.51), B (6.85) and control group (13.24). It was noticed that the TMA-N value of the marinated herring fillet decreased with increasing HPP level on all days. The results obtained revealed that the quality of the marinated herring fillet treated with HPP used in the research was effective during storage time. It was observed in one study that TMA-N quantity reduced in horse mackerel after HPP treatment at 330 MPa, for 10 minutes at 7°C and at 220 MPa, 250 MPa, 330 MPa, for 10 minutes at 25°C was lower than the control value (Erkan et al., 2011).

HPP treatment higher than 300 MPa gives seafood a very nimble cooked form (Hoover et al., 1989). TMA-N values reduced by stopping the protein breakdown through HPP treatment (Hernández-Andrés et al., 2005). It was also observed in another study of prawn that value of TMA-N increased in storage samples including control group (Basavakumar et al., 1998). It was observed that different HPP levels gave significantly different values of TMA-N during different days of storage ( $P < 0.05$ ) and these values were decrease as pressure was increased as in our study we observed that group G has highest pressure level and lowest TMA-N value.

## CONCLUSION

It is concluded from this article that due to perishable nature of fish, it needs proper treatment for long term preservation. There were many traditional thermal processing treatments, but they can change the taste and color of fish product. Therefore some advanced non thermal processing treatments should be applied to fish product preservation with minor change in taste, texture and color of final fish product, which is also a consumer requirement these days. From this study it can be concluded that high pressure processing treatment with combination of marination is an excellent approach to get good quality and taste of herring.

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**Table 1.** Changes in the physico-chemical properties of marinated herring fillets treated with HPP during storage at 4±1°C

	storage (days)	Control (A)	100MPa/5min (B)	100MPa/10min (C)	300MPa/5min (D)	300MPa/10min (E)	500MPa/5min (F)	500MPa/10min (G)
<b>pH</b>	<b>0</b>	4.40±0.02 <sup>Ag</sup>	4.38±0.01 <sup>Bg</sup>	4.36±0.01 <sup>Cg</sup>	4.35±0.01 <sup>Cf</sup>	4.32±0.01 <sup>Df</sup>	4.29±0.01 <sup>Eg</sup>	4.26±0.01 <sup>Fe</sup>
	<b>15</b>	4.43±0.01 <sup>Af</sup>	4.40±0.01 <sup>Bf</sup>	4.38±0.01 <sup>Cf</sup>	4.36±0.01 <sup>Df</sup>	4.33±0.01 <sup>Ef</sup>	4.31±0.01 <sup>Ff</sup>	4.27±0.01 <sup>Ge</sup>
	<b>30</b>	4.47±0.01 <sup>Ae</sup>	4.43±0.01 <sup>Be</sup>	4.40±0.01 <sup>Ce</sup>	4.39±0.01 <sup>Ce</sup>	4.35±0.01 <sup>De</sup>	4.33±0.01 <sup>Ee</sup>	4.29±0.01 <sup>Fd</sup>
	<b>45</b>	4.51±0.02 <sup>Ad</sup>	4.46±0.01 <sup>Bd</sup>	4.43±0.01 <sup>Cd</sup>	4.42±0.02 <sup>Cd</sup>	4.38±0.01 <sup>Dd</sup>	4.35±0.01 <sup>Ed</sup>	4.31±0.01 <sup>Fc</sup>
	<b>60</b>	4.55±0.01 <sup>Ac</sup>	4.50±0.01 <sup>Bc</sup>	4.47±0.01 <sup>Cc</sup>	4.44±0.01 <sup>Dc</sup>	4.41±0.01 <sup>Ec</sup>	4.38±0.01 <sup>Fc</sup>	4.34±0.01 <sup>Gb</sup>
	<b>75</b>	4.58±0.01 <sup>Ab</sup>	4.54±0.01 <sup>Bb</sup>	4.50±0.01 <sup>Cb</sup>	4.49±0.01 <sup>Cb</sup>	4.45±0.01 <sup>Db</sup>	4.40±0.01 <sup>Eb</sup>	4.34±0.01 <sup>Fb</sup>
	<b>90</b>	4.62±0.01 <sup>Aa</sup>	4.57±0.01 <sup>Ba</sup>	4.53±0.02 <sup>Ca</sup>	4.53±0.02 <sup>Ca</sup>	4.47±0.02 <sup>Da</sup>	4.43±0.03 <sup>Ea</sup>	4.37±0.01 <sup>Fa</sup>
<b>TMA-N (mg/100g)</b>	<b>0</b>	2.51±1.59 <sup>Ac</sup>	2.37±2.07 <sup>Ac</sup>	1.93±1.73 <sup>Aa</sup>	1.74±1.59 <sup>Ac</sup>	1.72±1.78 <sup>Ac</sup>	1.51±1.45 <sup>Ac</sup>	1.27±1.14 <sup>Ac</sup>
	<b>15</b>	2.94±1.23 <sup>Ac</sup>	2.76±0.98 <sup>Abc</sup>	2.50±1.90 <sup>Aa</sup>	2.03±1.49 <sup>Ac</sup>	1.69±1.03 <sup>Ac</sup>	1.73±1.35 <sup>Abc</sup>	1.54±1.93 <sup>Abc</sup>
	<b>30</b>	4.56±2.94 <sup>Abc</sup>	3.73±2.86 <sup>Aabc</sup>	3.60±2.61 <sup>Aa</sup>	3.29±2.08 <sup>Abc</sup>	3.42±2.43 <sup>Abc</sup>	3.36±2.37 <sup>Aabc</sup>	3.19±2.10 <sup>Aabc</sup>
	<b>45</b>	8.90±5.24 <sup>Aab</sup>	5.37±6.42 <sup>ABabc</sup>	5.09±6.75 <sup>ABa</sup>	4.13±1.32 <sup>Bab</sup>	3.85±1.98 <sup>Bab</sup>	3.80±1.77 <sup>Babc</sup>	3.45±1.06 <sup>Babc</sup>
	<b>60</b>	10.69±5.84 <sup>Aa</sup>	6.07±1.45 <sup>Bab</sup>	6.86±7.93 <sup>Aa</sup>	3.58±2.94 <sup>Bbc</sup>	3.91±2.39 <sup>Bab</sup>	3.95±2.89 <sup>Babc</sup>	3.42±1.82 <sup>Babc</sup>
	<b>75</b>	12.78±8.22 <sup>Aa</sup>	6.60±2.33 <sup>Ba</sup>	5.99±2.27 <sup>Ba</sup>	5.61±0.99 <sup>Ba</sup>	5.33±0.92 <sup>Ba</sup>	4.04±2.94 <sup>Bab</sup>	3.70±3.49 <sup>Bab</sup>
	<b>90</b>	13.24±5.77 <sup>Aa</sup>	6.85±1.96 <sup>Ba</sup>	6.51±2.48 <sup>Ba</sup>	5.13±0.84 <sup>Bab</sup>	5.62±0.19 <sup>Ba</sup>	5.29±1.97 <sup>Ba</sup>	4.25±2.44 <sup>Ba</sup>
<b>TVB-N (mg N/100 g)</b>	<b>0</b>	17.60±0.03 <sup>Ad</sup>	15.85±0.03 <sup>Ad</sup>	16.09±0.03 <sup>Ad</sup>	16.17±0.03 <sup>Ad</sup>	16.32±0.02 <sup>Ac</sup>	15.33±0.03 <sup>Ae</sup>	16.05±0.03 <sup>Ac</sup>
	<b>15</b>	25.08±0.03 <sup>Acd</sup>	19.90±0.02 <sup>Bd</sup>	18.56±0.02 <sup>Bd</sup>	19.42±0.02 <sup>Bd</sup>	18.82±0.03 <sup>Bc</sup>	17.17±0.03 <sup>Bde</sup>	16.30±0.02 <sup>Bc</sup>
	<b>30</b>	33.64±0.03 <sup>Abc</sup>	27.93±0.03 <sup>ABcd</sup>	25.53±0.02 <sup>ABCcd</sup>	26.08±0.03 <sup>ABCcd</sup>	24.48±0.04 <sup>ABCbc</sup>	21.79±0.03 <sup>BCcd</sup>	17.67±0.03 <sup>Cc</sup>
	<b>45</b>	39.81±0.03 <sup>Ab</sup>	37.66±0.02 <sup>ABac</sup>	33.74±0.03 <sup>ABbc</sup>	34.24±0.02 <sup>ABbc</sup>	35.06±0.02 <sup>ABab</sup>	24.65±0.02 <sup>BCbc</sup>	20.75±0.02 <sup>Cbc</sup>
	<b>60</b>	44.31±0.02 <sup>Ab</sup>	40.89±0.02 <sup>Aab</sup>	38.34±0.03 <sup>ABab</sup>	37.57±0.01 <sup>ABab</sup>	34.00±0.03 <sup>ABCa</sup>	26.94±0.03 <sup>BCab</sup>	24.39±0.03 <sup>Cab</sup>
	<b>75</b>	58.37±0.03 <sup>Aa</sup>	48.56±0.01 <sup>ABab</sup>	45.66±0.02 <sup>Bab</sup>	41.77±0.02 <sup>Bab</sup>	38.05±0.03 <sup>BCa</sup>	28.52±0.02 <sup>Cab</sup>	27.32±0.02 <sup>Ca</sup>
	<b>90</b>	56.70±0.04 <sup>Aa</sup>	51.91±0.02 <sup>ABa</sup>	42.94±0.03 <sup>BCa</sup>	47.91±0.03 <sup>ABa</sup>	43.52±0.01 <sup>BCa</sup>	31.76±0.03 <sup>CDa</sup>	28.40±0.02 <sup>Da</sup>

Means indicated by different capital letters in the same row differ significantly ( $p < 0.05$ ). Means indicated by different lowercase letters in the same column differ significantly ( $p < 0.05$ ).