

# RADICAL FORMATION, OXIDATIVE DETERIORATION AND ITS PREVENTION BY ANTIOXIDANT ACTIVE PACKAGING: A CASE STUDY OF MEAT PROCESSED BY HIGH PRESSURE PROCESSING

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## Abstract

High pressure processing (HPP) has been proved to be an effective processing technology to extend the shelf life of fresh meats by microbial inactivation with minimal effects on nutritional value. However, oxidative reactions in meats can often lead to product deterioration even though microbial growth is under control. In meats, HPP induces the formation of free radicals an early event that occurs prior to the progression of oxidation. The present paper describes the formation of free radicals in chicken breast either by heat or high pressure processing treatments. The possibility of delaying the microbial growth and the oxidative reactions by combining high pressure treatment with antioxidant active packaging is presented as an alternative food preservation method for a shelf-life extension.

Key words: High pressure processing, radical formation, meat, oxidation, antioxidant active packaging

## 1. Introduction

Microbial growth and oxidative reactions are the most frequent causes of meat spoilage. Preservation methods must be implemented to minimise the losses in the processing and distribution by means of preservation technologies which protect the food from microbial or chemical deterioration.

High pressure processing (HPP) is a non-thermal processing technology which is becoming of increasing importance in the production of minimally processed foods for which shelf-life can be extended primarily through inactivation of bacteria, spoilage microorganisms and food borne pathogens (such as *Listeria* and *Salmonella*) by applying pressures above 400 MPa and with minimum loss of nutritional value and sensory quality.

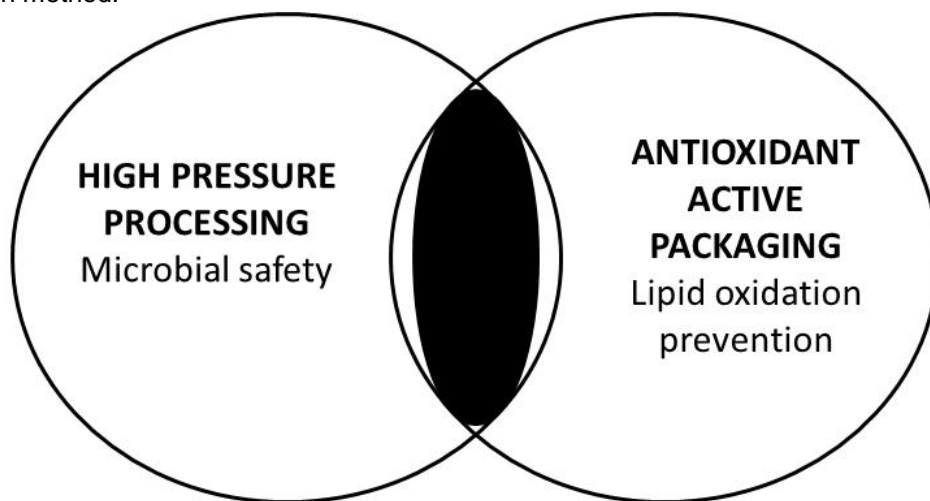
HPP of meat triggers lipid oxidation which reduces shelf-life and affects the flavour of the product. It has been established that a pressure above 300-400 MPa is critical for inducing lipid oxidation (Orlien et al., 2000, Beltran et al., 2004). Nevertheless, the lipid oxidation can be prevented by using antioxidants. Rosemary extract has been successfully employed to protect chicken meat processed by HP (Bragagnolo et al., 2007).

The oxidative stability of meat is a complex phenomenon which depends on (1) compositional factors such as lipid content, fatty acids profile and components with antioxidant (e.g. vitamin E and others constituents) or reducing (e.g. vitamin C) functionality and pro-oxidants (e.g. Fe and Cu) as well as (2) external conditions such as the deboning method, the chilling rate, the processing (heating, mincing, and addition of antioxidants or curing agents), and the exposure to light and oxygen, and the temperature conditions (Skibsted et al. 1998).

Active packaging is designed to deliberately incorporate material components in the packaging that release or absorb substances from or into the packaged food or the surrounding environment in order to extend the shelf-life and maintain or improve the condition of packaged food. The technology provides several advantages compared to direct addition, such as less amount of active substances requirement and localization of the activity to the surface (Coma, 2008). The European Food Authorities have recently passed new legislation in order to regulate the use of active substances in packaging intended to come into contact with food (EC, May 2009).

The present paper describes the challenge of using HPP to maintain freshness by microbial inactivation and the particular activity of antioxidants in active packaging to efficiently protect against lipid oxidation. The use of HPP in combination with antioxidant active packaging represents a novel food processing technology (Figure 1).

**Figure 1.** Combination of high pressure processing with antioxidant active packaging as a novel preservation method.



## 2. Materials and Methods

### 2.1. Meat model system

The meat model system was chicken breast purchased from a local supermarket and added with the spin-trap, N-t-butyl- $\alpha$ -phenyl nitron (PBN), as described in Bolumar et al. (2011). Aliquots of 4.5 ml of the meat slurries were vacuum packed in plastic bags (PA/PE 20/70, 32 oxygen cm<sup>3</sup>/ m<sup>2</sup> d bar at 23 °C and 75 % RH, SFK, Hvidovre, Denmark) at 99 % vacuum for further processing.

### 2.2. Heat treatment and high pressure treatment

The meat slurries were submitted to incubation at 55, 65, and 75 °C in a water bath or processed by HPP in a Food Processing Press (QFP-6 Avure Technologies AB, Vasterås, Sweden) at 500, 600, and 700 MPa at 25 °C over a period of time between 0 and 60 min (Bolumar et al. 2012). Samples were taken at 0, 10, 30 and 60 min for quantification of formed-radicals.

### 2.3. Free radical quantification by electron spin resonance (ESR) spectroscopy

The ESR measurements were made in a JEOL FR 30 ESR spectrophotometer (JEOL Ltd., Tokyo, Japan) at room temperature following procedure described in Bolumar et al. (2012).

### 2.4 Chicken breast and chicken thigh patties preparation and antioxidant active packaging

Chicken breast and chicken thigh patties and antioxidant active packaging containing rosemary was prepared according to Bolumar et al. (2011).

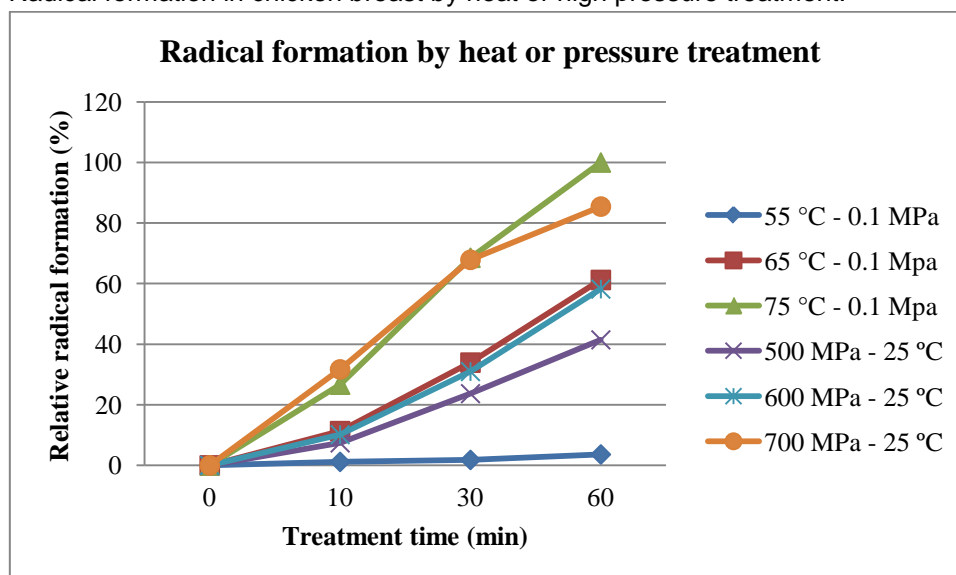
### 2.5. Lipid oxidation quantification

The extent of lipid oxidation was measured by the TBARS method described by Jensen, Skibsted, Jacobsen & Bertelsen (1995).

### 3. Results and discussion

Application of heat or pressure leads to formation of radicals in meat model systems (Figure 2). According to the effect of the processing conditions on the reaction rate, three groups of increasing order of radical formation were established: (1) 55 °C at 0.1 MPa, (2) 500 and 600 MPa at 25°C and 65 °C at 0.1 MPa, and (3) 700 MPa at 25 °C and 75 °C at 0.1 MP.

**Figure 2.** Radical formation in chicken breast by heat or high pressure treatment.



A free radical is easily formed when a covalent bond is broken and one electron remains with each newly formed atom. However, according to the thermodynamic principles the effect of pressure on covalent bonds can be almost neglected under regular pressures ranging from 0.1 to 1500 MPa and low temperatures (0-40 °C) (Aertsen et al., 2009). However, radicals also formed at high pressure processing conditions. The mechanisms behind are unknown but must be associated to couple electron transfer or redox reaction favoured under high pressure conditions.

Heating can promote lipid peroxidation by similar mechanisms as those proposed for high pressure processing: disruption of muscle cell, inactivation of antioxidant enzymes, and release of iron from myoglobin (Min & Ahn, 2005). In contrast to temperature, pressure is transmitted instantaneously to the entire food system, so no gradients takes place and less time is needed to reach the required pressure in the centre of the product. Pressure and temperature applied and treatment duration are main factors determining the amount of radical formed during the preservation treatment. The higher the pressure, the temperature and the duration of the treatment the higher the formation is. Hence the use of antioxidant active packaging could be an alternative method to restrain the lipid oxidation in meats prone to lipid oxidation.

**Table 1.** Lipid oxidation measured as TBARs ( $\mu\text{mol}/\text{kg}$  dry meat) (mean  $\pm$  standard deviation) at the surface and the inner part of chicken breast and thigh patties submitted to high pressure and packaged in antioxidant active packaging containing rosemary.

	Chicken breast patties		Chicken thigh patties	
	day 11	Day 25	day 11	day 25
Control – surface	21.7 $\pm$ 2.2	22.0 $\pm$ 0.8	47.1 $\pm$ 0.8	44.9 $\pm$ 1.3
Control – inner part	9.9 $\pm$ 0.5	18.5 $\pm$ 0.3	20.9 $\pm$ 0.4	28.3 $\pm$ 0.1
Antioxidant active packaging – surface	12.6 $\pm$ 1.3	18.6 $\pm$ 0.5	14.4 $\pm$ 1.1	16.5 $\pm$ 0.3
Antioxidant active packaging – inner part surface	8.8 $\pm$ 0.2	11.5 $\pm$ 0.7	13.6 $\pm$ 0.5	14.0 $\pm$ 0.3

In the Table 1 can be visualized the oxidation observed at the surface and the inner part during the storage of chicken breast and thigh patties submitted to high pressure and packaged in antioxidant active packaging containing rosemary. In brief, the lipid oxidation was higher at the surface of the meat and the active packaging was able to restrain lipid oxidation at the surface as well as in the inner part. This effect was more outstanding in chicken thigh patties as the level of oxidation was higher due to the more prone nature of thigh to oxidation compared to chicken breast (oxidative metabolism and higher fat content).

## Conclusion

Radical formation by heat or pressure treatments is an early event in lipid oxidation prior to progression of lipid oxidation. HPP-induced lipid oxidation in chicken meats can be prevented by the use of antioxidant active packaging. The combined usage of HPP with antioxidant active packaging is a promising method to extend the shelf life by limiting microbial growth and oxidative reactions. Further research is needed particularly on the mechanisms behind the formation of radicals under pressure conditions and on the development of antioxidant active films suitable for industrial applications.

## References

1. Aertsen, A., Meersman, F., Hendrickx, M., E.G. Vogel, R.F., & Michiels, C.W. (2009). Biotechnology under high pressure: applications and implications. *Trends in Biotech.* 27, 434-441.
2. Beltran, E., Pla, R., Yuste, J., & Mor-Mur, M. (2004). Use of antioxidants to minimize rancidity in pressurized and cooked chicken slurries. *Meat Sci.*, 66, 719-725.
3. Bolumar, T., Andersen, M.L., & Orlien, V. (2011). Antioxidant active packaging for chicken meat processed by high pressure treatment. *Food Chemistry*, 129, 1406-1412.
4. Bolumar, T., Skibsted, L.H., & Orlien, V. (2012). Kinetics of the formation of radicals in chicken breast during high pressure processing. *Food Chemistry*, 134, 2114–2120.
5. Bragagnolo, N., Danielsen, B., & Skibsted, L.H. (2007). Rosemary as antioxidant in pressure processed chicken during subsequent cooking as evaluated by electron spin resonance spectroscopy. *Innov. Food Sci. Emer. Technol.*, 8, 24–29.
6. Coma, V. (2008). Bioactive technologies for extended shelf life of meat-based products. *Meat Science*, 78: 90-103.
7. EC, European Commission. (2009). Commission Regulation (EC) No 450/2009 of 29 May 2009 on active and intelligent materials and articles intended to come into contact with food. *Official Journal of the European Union*, L 135: 3-11.
8. Jensen, C., Skibsted L.H., Jakobsen, K., & Bertelsen., G. (1995). Supplementation of broiler diets with all-rac-alpha – or a mixture of natural sources RRR-alpha –, gamma –, delta – tocopheryl acetate. II: Effect on the oxidative stability of raw and precooked broiler meat products. *Poultry Sci.*, 74, 2048-2056.
9. Ma, H.J., Ledward, D.A., Zamri, A.I., Frazier, R.A., & Zhou, G.H. (2007). Effects of high/pressure thermal treatment on lipid oxidation in beef and chicken muscle. *Food Chemistry*, 104, 1575-1579.
10. Min, B., & Ahn, D.U. (2005). Mechanism of lipid peroxidation in meat and meat products – A review. *Food Science Biotechnology*, 14, 152-163.
11. Orlien, V., Hansen, E., Skibsted, L.H. (2000). Lipid oxidation in high-pressure processed chicken breast muscle during chill storage: critical working pressure in relation to oxidation mechanism. *Eur. Food Res. Technol.*, 211: 99-104.
12. Skibsted, L.H., Mikkelsen, A., & Bertelsen, G. (1998). Lipid-derived off-flavours in meat. In Shahidi, F. (Eds.), *Flavor of Meat, Meat Products and Seafoods* (2nd Edition) (pp. 217-256). New York, Springer – Verlag.