Contents lists available at SciVerse ScienceDirect

Food and Bioproducts Processing

journal homepage: www.elsevier.com/locate/fbp

Antioxidant protection of high-pressure processed minced chicken meat by industrial tomato products

A.B. Alves[a], N. Bragagnolo[a], M.G. da Silva[b], L.H. Skibsted[c], V. Orlien[c,*]

[a] Department of Food Science, State University of Campinas, P.O. Box 6121, 13083-862 Campinas, SP, Brazil
[b] Food Science and Quality Center, Institute of Food Technology, 13070-178 Campinas, SP, Brazil
[c] Food Chemistry, Department of Food Science, Faculty of Life Sciences, University of Copenhagen, Rolighedsvej 30, DK-1958 Frederiksberg C, Denmark

ABSTRACT

Pressure-treatment at 300 MPa of minced chicken breast meat did not induce significant lipid oxidation during chill storage for up to 15 days, while pressure-treatment at 600 and 800 MPa enhanced formation of secondary lipid oxidation products measured as thiobarbituric acid reactive substances. However, a waste product from industrial tomato paste production was found to yield efficient protection against lipid oxidation in pressurized chicken meat. Addition of 0.30% tomato waste or of 0.10% final tomato paste to minced meat led to a lag phase of 6 days for formation of secondary oxidation products in meat pressure treated at 600 MPa. The waste product seemed special efficient in protecting chicken meat pressurized at 800 MPa, as a notably lower rate of formation of secondary oxidation products was found. Flavonoids washed out with the waste fraction may be more efficient as antioxidant than the other phenolics or carotenoids present in the other paste processing fractions. Addition of tomato paste fractions did not influence radical formation measured by electron spin resonance spectroscopy and the tomato waste is considered to be effective as antioxidant at subsequent reactions leading to secondary lipid oxidation products.

Keywords: High pressure; Chicken meat; Tomato paste products; Lipid oxidation; ESR; Flavonoids

1. Introduction

High-pressure processed chicken meat seems to have a market potential as a minimal processed meat product due to less lipid oxidation resulting from pressure processing compared to thermal processing and due to less pressure-induced discoloration of chicken meat compared to pork and beef muscle. Previous studies have shown that lipid oxidation in vacuum-packed chicken meat is initiated during high pressure treatment due to the relatively high content of unsaturated fatty acids in poultry fat, even if the meat was vacuum-packed (Orlien et al., 2000; Beltran et al., 2004; Wiggers et al., 2004; Bragagnolo et al., 2005; Mariutti et al., 2008). It is well-documented that the use of natural antioxidants from herbs and spices in meat products may prevent or delay lipid oxidation. Especially, rosemary and sage were found to be effective antioxidants in high-pressure treated chicken meat during chill storage (Bragagnolo et al., 2005; Mariutti et al., 2008). However, it was found that garlic showed prooxidative effects in chicken meat treated at pressures above 300 MPa (Mariutti et al., 2008). Moreover, both sage and garlic were found to act as radical scavengers in an isolated chicken lipid phase, while sage acted as a prooxidant and garlic showed no effect in an isolated chicken aqueous phase (Mariutti et al., 2008). This remarkable result calls for a detailed evaluation of the effect of any potential antioxidant on the progression of lipid oxidation in the real food product prior to practical use. Tomato is rich in antioxidants like carotenoids, flavonoids, and vitamin E and C, and a vegetable often used in meat dishes. Studies have shown that high pressure treatments of tomato puree did not affect carotenoids content or antioxidant activity of hydrophilic and lipophilic fractions (Hsu, 2008; Garcia et al., 2006; Sánchez-Moreno et al., 2006). Another study showed that high pressure treatment of onions would maintain or increase the levels of total phenol and flavonols (Roldán-Marín et al., 2009). In order to evaluate tomato products as a source of

* *Corresponding author*. Tel.: +45 35333226; fax: +45 35333344.
E-mail addresses: drialves@ital.sp.gov.br (A.B. Alves), vor@life.ku.dk (V. Orlien).
Received 31 March 2011; Received in revised form 17 October 2011; Accepted 25 October 2011
0960-3085/$ – see front matter © 2011 The Institution of Chemical Engineers. Published by Elsevier B.V. All rights reserved.
doi:10.1016/j.fbp.2011.10.004

natural antioxidants to be used for protection of pressure treated chicken meat during subsequent storage, the effect of working pressure on lipid oxidation in chicken meat without or with various industrial tomato paste processing fractions added, including waste, was investigated. For this investigation three scenarios was chosen; low pressure, 300 MPa, to have a 'stable' sample, intermediate pressure, 600 MPa, to be just above the critical pressure, and high pressure, 800 MPa, as a worst case scenario in order to induce lipid oxidation.

2. Materials and methods

2.1. Chemicals and standards

Ethanol, methanol, acetonitrile, acetic acid, and ethyl acetate was of HPLC grade. Gallic acid, rutin, naringenin, lycopene and beta-carotene were purchased from Sigma (St. Louis, MO, USA), quercetin-rhamnoside (quercitrin) was obtained from Extrasynthese (Z.I. Lyon Nord, Genay, France). Malonaldehyde was from Merck (Merck-Schchardt, Dr. Th. Schchardt & Co., D-85662 Hohenbrunn) and 1,1,3,3-tetraethoxypropane (TEP) was from Merck (Darmstadt, Germany). α-Tocopherol was from Merck (Darmstadt, Germany) and γ-tocopherol was from Sigma–Aldrich Chemie (Steinheim, Germany). All standards used were purchased with minimum purity of 95% and all other reagents were of analytical grade.

2.2. Tomato samples

The various tomato products were collected at Predilecta Alimentos Ltda (Matão, São Paulo, Brazil) in September of 2006. The products were collected at 4 different sites of the tomato paste processing line and accordingly labeled: in natura, finishing, final, and waste (Fig. 1). Each processing step was sampled 3 times during a normal processing day with 3 h of interval between each sample. The samples were pooled totalizing 4 kg of in natura product, 4 kg of finishing product, 2 kg of final product, and 1 kg of waste product. Immediately after collection, the products were frozen on dry ice followed by freeze-drying and storing at −20 °C until use. It was decided to pool the collections from the tomato processing line in order to focus on the difference between processing and to minimize the effect of variation between the individual tomato fractions.

2.3. Total phenolic content

The tomato products were characterized with respect to total phenolics. The phenolic extracts were obtained as follows: 50 mL of the extraction solution (60% ethanol with 0.1% acetic acid) was added to 1.5 g of freeze-dried tomato sample. The mixture was homogenized for 1.5 min in an Ultra turrax® (Ika, Junke & Kunkel, Germany) and filtrated through a filter paper to a 250 mL round bottom flask. The extraction was repeated twice and the filtrates were added to the same flask. The combined extract was concentrated on a rotary evaporator (Rotavapor R-144, Büchi, Flawil, Switzerland) with a vacuum pump (Julabo F25, Seelbach, Germany) in a water bath (Waterbath B-840, Büchi) set at 45 °C. The concentrate was transferred to a 50 mL volumetric flask and the volume was made with milli-Q water (milli-Q ultra pure system, USA). Total phenolic content was determined using Singleton and Rossi (1965) method adapted by Kim et al. (2003). Measurements were carried out in a UV–vis spectrophotometer Varian Cary 50 (Varian Australia Pty Ltd., Mulgrave, Victoria, Australia) at 750 nm against a blank of reagents. Analysis was performed in triplicate from extraction. Gallic acid was used as standard and the results were expressed as gallic acid equivalents (in mg gallic acid equivalents/100 g freeze-dried products).

2.4. Flavonoids analysis

For the flavonoids analysis the previous extract was concentrated and purified by solid phase extraction (SPE): 20 mL of the phenolic extract was applied in a polyamide column (1 g/6 mL) Chromabond® (Macherey-Nagel, Düren, Germany) pre-conditioned with 20 mL of methanol and 60 mL of water. The column was washed with 20 mL of water and the flavonoids fraction was eluted with 50 mL of methanol. The methanol was evaporated on a rotary evaporator at 45 °C and the extract was dissolved in 5 mL of methanol. The purified extract was used to determine total flavonoids and for flavonoids analysis by high performance liquid chromatography (HPLC). Total flavonoid content was determined using the method of Zishen et al. (1999). Measurements were carried out in a UV–vis spectrophotometer Varian Cary 50 at 510 nm against a blank of reagents. Analysis was performed in triplicate from extraction. Rutin was used as standard and the results were expressed as rutin equivalents (in mg rutin equivalents/100 g freeze-dried products). For HPLC analysis the extract was filtered through 0.45 μm membrane and injected in to a HPLC system (Shimadzu, model 10Avp, Kyoto, Japan) equipped with a quaternary pump (LC-10ATvp), a column heater (CTO-10ASvp), a diode array detector (DAD-UV–vis) (SPD-M10Avp), and a Rheodyne® injector with a 20 μL sample loop. Separation was carried out at 20 °C using an Inertsil ODS-3 column (250 mm × 4.6 mm, 5 μm, GL Sciences, Tokyo, Japan), mobile phase composed by 0.1% acetic acid solution (A) and acetonitrile:methanol (96:4, v/v) (B) at a flow rate of 1 mL/min. Detection was made at 290 nm for naringenin, and at 365 nm for rutin and quercitrin. Quantification was made using external calibration curves. Analysis was performed in triplicate and the results were expressed in mg/100 g of freeze-dried sample (dry matter basis).

2.5. Carotenoid content

Lycopene and β-carotene contents of tomato products were determined using a validated method for tomato products describe by Alves et al. (2010). HyfloSupercel was added to 0.5 g of freeze-dried tomato sample, homogenized and hydrated with 20 mL of water. Carotenoids were extracted once with 50 mL of acetone and then three times with 30 mL of acetone for 1 min using an Ultra turrax® (Ika, Junke & Kunkel, Germany). 20 mL of acetone extract were transferred to a separation funnel containing 50 mL of petroleum ether. Acetone was removed with water and a new acetone aliquot was added. The process was repeated until all acetone extract was transferred and washed. Petroleum ether with carotenoids was collected in a 50 mL volumetric flask and the volume was made up with petroleum ether. For β-carotene quantification, 1 mL of the extract was dried under nitrogen and dissolved in 1 mL of mobile phase. For lycopene, 0.5 mL of extract was dried and dissolved in 5 mL of mobile phase. Carotenoids were analyzed using a liquid chromatograph (Shimadzu, model 10, Kyoto, Japan) composed by a binary pump (Shimadzu LC-10, Kyoto, Japan) and diode array detector (DAD-UV–vis) (Shimadzu SPD-M10A, Kyoto, Japan). Separation was carried

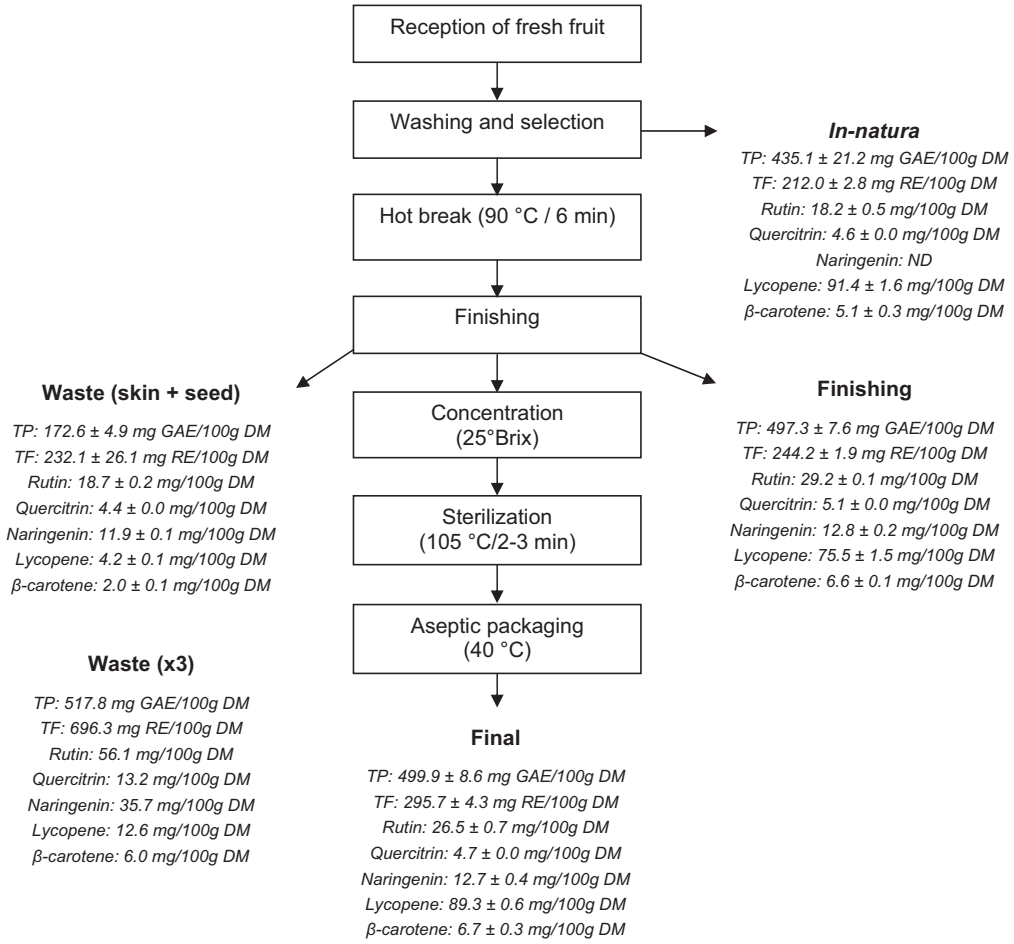

Fig. 1 – The tomato paste processing scheme showing the 4 different sites of collecting the tomato products with the total phenolic content (TP), total flavonoids content (TF), rutin, quercitrin, naringenin, lycopene, and β-carotene content to be used in high-pressure processed chicken meat. DM = dry matter.

out using an Lichrospher® 100 RP18 column (125 mm × 4 mm, 5 μm, Merck, Germany), and mobile phase composed by acetonitrile:methanol:ethyl acetate (73:20:7, v/v/v) at 1.5 mL/min isocratic flow. Detection was done at 450 nm and quantification was made using external standards at 0.9 μg/mL for β-carotene and 2.0 μg/mL for lycopene. Analysis was performed in duplicate.

2.6. *Preparation of chicken meat samples*

Frozen chicken breast was obtained from Rose Poultry A/S (Vinderup, Denmark) and was thawed for 48 h at 5 °C. The chicken breasts were minced in an electric mincer after removing connective and adipose tissues. All the minced meat was manually mixed and divided into five portions of 1.34 kg each. The first portion (S1) was the control with no addition of tomato products; the second portion (S2) was mixed with 0.10% (w/w) in natura tomato sample; the third one (S3) was mixed with 0.10% (w/w) finishing product; the fourth portion (S4) was mixed with 0.10% (w/w) final product; and the last one (S5) was mixed with 0.30% (w/w) waste product as this product had around 3 times less phenolics and 20 times less lycopene than the average of the others tomato products. Each chicken portion was well homogenized with a fork to incorporate the tomato products and sequentially divided in 20 polyethylene bags containing two portions of 10 g each, vacuum-packed and stored at 5 °C for 1 day (S1, S2, S3) or 2 days (S4, S5) before high-pressure processing.

2.7. *High-pressure processing*

The vacuum-packed meat samples were submerged in the pressurizing chamber of a QUINTUS Food Processing Cold Isostatic Press QFP-6 (Avure Technologies AB, Västerås, Sweden) with water thermostated at 10 °C as the pressure transmission fluid. The meat samples were treated at 300, 600, or 800 MPa hydrostatic pressures for 10 min (20 bags of each chicken sample were treated at each pressure in two batches of 10 bags each). During high-pressure treatment, the temperatures in the pressure chamber increased and the maximum temperatures reached were 16.1 °C at 300 MPa, 29.1 °C at 600 MPa, and 43.4 °C at 800 MPa, as measured by a built-in thermocouple. The depressurizing was initiated automatically after 10 min and instantly resulted in a decrease in temperature to approximately 5, 0, and −2 °C after working at 300, 600, and 800 MPa, respectively. The temperature fluctuation in the meat samples will be quite similar to the temperature increase during pressure build-up and temperature decrease during depressurization.

2.8. *Storage*

Immediately after high-pressure processing all 20 bags of each combination of tomato products and pressure treatment were opened and closed again to permit air entrance to present worst-case-scenario and then stored at 5 °C in the dark. Samples (2 bags, each one containing two meat portions) were

taken for analysis of color, ESR, and TBARS on days 0, 3, 6, 8, 10, 13, and 15. Vitamin E was analyzed on samples from days 0 and 15. Raw sample, without addition of any tomato products and not subjected to high-pressure treatment, were also analyzed.

2.9. *Electron spin resonance spectroscopy*

The relative radical content was measured by electron spin resonance (ESR) spectroscopy as described by Nissen et al. (2000). Samples were freeze-dried at −40 °C in an Edwards Modulyo Freeze-Drier (Edwards High Vaccum, Britain) in order to trap radicals in the rigid structure of the low water-activity product. The freeze-dried samples were ground in an A10 Janke & Kunkel Mill (Janke & Kunkel GmbH & Co. KG – IKA-Labortec, Staufen-Br, Germany) with water refrigeration for 7 cycles of 15 s each, until a fine chicken meat powder was obtained. Between 0.3 and 0.6 g of sample (accurately weighed) was transferred to a cylindrical, thin-walled 702-PQ-7 clear-fused quartz (CFQ) tube (o.d., 5 mm; Wilmad Glass Company Inc., Buena, NJ) which was gently tapped against the table in order to establish a dense and uniform packing (column height, approximately 6 cm; density, approximately 0.07 g/cm, calculated for each individual meat sample). The ESR measurements were performed using a JES FR30 Free Radical Monitor (JEOL, Tokyo, Japan) with the following parameters: sweep time 1 min, sweep width 7.5 mT, microwave power 4 mW, modulation width 0.63 mT, and time constant 0.1 s. The amount of radicals was quantified as the height of the ESR signals relative to the signals from a built-in manganese standard and normalized by the density (g/cm) of the meat sample in the ESR tube. Results are expressed as mean of two different samples submitted to the same treatment. Relative Standard Deviation (RSD) was around 10%.

2.10. *Analysis of thiobarbituric acid reactive substances*

The thiobarbituric acid reactive substances (TBARS) were determined in 5.00 ± 0.10 g of homogenized meat samples according to Sørensen and Jørgensen (1996). TBARS were expressed in μmol malonaldehyde (MDA) per kg meat. A standard curve between 0.1 and 12.0 μmo/L of TEP was used. The results presented are the means of measurements on two different samples exposed to the same treatment. RSD was around 6%.

2.11. *Vitamin E determination*

Analysis of α- and γ-tocopherol content in chicken meat were performed as previously described (Jensen et al., 1997). Vitamin E was expressed as mg of α- and γ-tocopherol per kg of meat using an external standard curve (concentration range from 0.5 to 10 mg/L) for α- and γ-tocopherol, respectively. The reported values are means of duplicate measurements each on two different samples exposed to the same treatment.

2.12. *Color measurement*

The color measurements were performed using a Color-Guide 45/0 (Byk-Gardner, Geretsried, Germany). The L^*, a^* and b^* values were measured three times on the surface of meat samples processed in duplicate (Illuminant D65/10°).

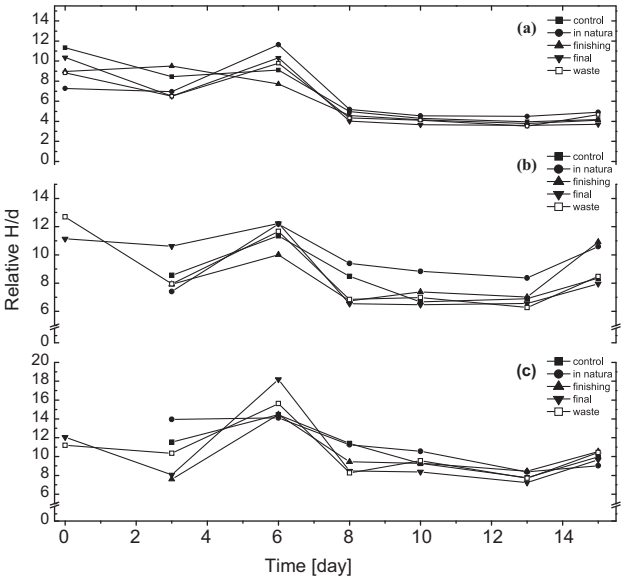

Fig. 2 – Formation of radicals (H, the relative height of the ESR signal normalized by the density) in freeze-dried pressurized minced breast chicken meat with and without addition of tomato products during subsequent chill storage in the dark: (a) 300 MPa (b) 600 MPa, and (c) 800 MPa.

2.13. *Statistical analysis*

The results were submitted to analyses of variance and the means were classified by LSD ("least square difference" at $p < 0.05$). The adjusted means were calculated by multiple regression by the General Linear Model (GLM) procedure. All the calculations were performed using the software Statistica for Windows 5.5 (StatSoft Inc., Tulsa, USA).

3. Results and discussion

The production of tomato paste includes several processing steps, some of them at high temperatures, which may change both the total phenolic content and the distribution between specific plant phenols in the final product compared to the raw material. Accordingly, the antioxidant activity of the various products and waste fraction with respect to lipid oxidation in a real meat product is expected to vary. The tomato waste fraction differs from the tomato products not only in the content of total phenolic compounds but also in content of specific flavonoids, lycopene, and β-carotene (Fig. 1). However, addition of 3 times as much waste product to the meat, as done in the present investigation, results in comparable total phenolic and β-carotene contents, higher flavonoids contents, but still less lycopene content.

The potential of the different tomato products to diminish the pressure-induced lipid oxidation in chicken breast meat during chill storage at 5 °C for up to 15 days was quantified as the formation of radicals using electron spin resonance spectroscopy (ESR). Fig. 2 shows that approximately similar levels of radicals were obtained immediately after pressure treatment (Day 0), indicating that high pressure treatment initiated lipid oxidation to the same extent irrespective of pressure level and addition of tomato products to the meat. The radical formation is an early event in lipid oxidation and, thus, a predecessor for the further development of lipid oxidation resulting in lipid hydroperoxides and secondary lipid oxidation products. Due to the autocatalytic nature of the

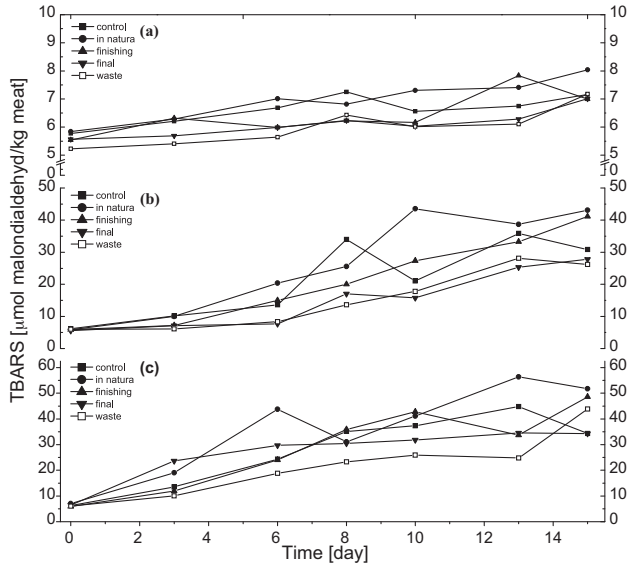

Fig. 3 – Formation of secondary lipid oxidation products (TBARS) in pressurized minced chicken breast meat with and without addition of tomato products during subsequent chill storage in the dark: (a) 300 MPa (b) 600 MPa, and (c) 800 MPa.

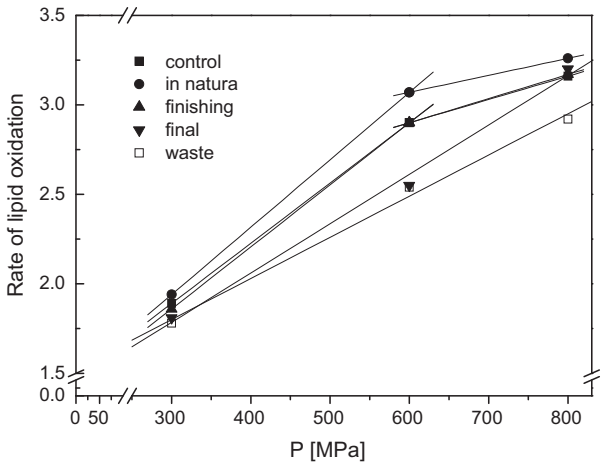

Fig. 4 – Rate of development of lipid oxidation expressed as ln(TBARS/t) (adjusted means of TBARS/t in μmol kg^{-1} day^{-1}) as a function of working pressure.

progression phase in the overall lipid oxidation process, an increase in radical formation as a result of high-pressure treatment was expected. However, a decline in the content of radicals during storage was observed for the meat pressurized at 300 MPa (Fig. 2(a)), indicating that the radicals formed during pressure treatment is scavenged and may not enhance lipid oxidation further. The steady-state level of radical content at 600 and 800 MPa (Fig. 2(b) and (c)) indicates a continuous formation and scavenging of radicals. Surprisingly, no difference in the development of radical formation between meat with or without addition of tomato products is noted within the same pressure treatment as can be seen in Fig. 2. Apparently, the compounds present in the tomato products does not limit the radical formation, although radical scavenging has been identified as important for the protective effect of antioxidants against lipid oxidation. In addition, rosemary has previously been shown to inhibit lipid oxidation in chicken meat processed at 600 MPa by lowering radical formation as shown by ESR spectroscopy (Bragagnolo et al., 2007).

The effect of the different tomato products on the further progression of lipid oxidation in high pressure treated chicken meat during chill storage at 5 °C for up to 15 days was investigated using TBARS as a measure of the extent of lipid oxidation. Initially, after pressure treatment (Day 0), the extent of lipid oxidation is the same, but during storage the extent of lipid oxidation was found to vary with the pressure level as seen from the developments in the TBARS curves (Fig. 3). Similar levels of TBARS-values and developments in lipid oxidation in pressure-treated chicken breast meat were found by Mariutti et al. (2008). From the TBARS-values it is directly seen that pressure at 300 MPa did not induce significant lipid oxidation during the entire storage period for chicken meat, as the level of TBARS was below 8 μmol/kg for all meat samples (Fig. 3(a)), in agreement with the ESR results. TBARS values above 15 are sensorially unacceptable for most meat products (Mielche and Bertelsen, 1995). In contrast, pressure treatments at 600 and 800 MPa had a prooxidative effect on chicken meat, as lipid oxidation was observed to increase significantly during storage (Fig. 3(b) and (c)). A critical working pressure for oxidative damage seems to exist between 300 and 600 MPa in agreement with previous findings (Orlien et al., 2000; Beltran et al., 2003, 2004; Mariutti et al., 2008). From the developments of the secondary lipid oxidation products as indicated by the TBARS-values, it is seen that the final product (S4) and the waste product (S5) had some effect in protecting chicken meat against pressure-induced lipid oxidation. At 300 MPa it is seen that during storage for 13 days, the development of TBARS for the samples S4 and S5 was lower than the control (S1) (Fig. 3(a)). For meat pressurized at 600 MPa a lag phase of 6 days was identified for chicken meat with the two most efficient tomato products added (waste and final products) while no lag phase was observed for the control sample. Accordingly after 6 days of storage, the samples S4 and S5 were still sensorially acceptable with TBARS-values around 8 μM/kg, while S1 were very close to the limit of 15 μM/kg with an actual TBARS-value of 13.6 μM/kg (Fig. 3(b)). Moreover, during the entire chill storage period, the development of lipid oxidation for these samples was significantly lower than the control sample (Fig. 4). For 800 MPa pressure treatment, lipid oxidation is efficiently induced in chicken meat and develops rapidly, although a lower rate of development of secondary oxidation products clearly was observed for the meat with the waste product added (S5) (Figs. 3(c) and 4). Notably, lycopene, which otherwise is known as a very efficient singlet oxygen quencher and radical scavenger (Mortensen and Skibsted, 1997), seems to be of little importance in protection against pressure induced lipid oxidation. The tomato product waste with the lowest lycopene content, showed the most protective effect against lipid oxidation. Specific flavonoids washed out with the waste fraction, like rutin, quercitrin, and naringenin, may be more efficient as antioxidant than carotenoids with respect to preventing lipid oxidation in pressurized chicken meat. This observation is considered to be of practical importance to the meat processing industry.

An assessment of the effect of the tomato samples on the progression of lipid oxidation in pressure-treated chicken breast meat emerged from the statistical analysis based on the adjusted means of the ln(TBARS)-values (Fig. 4). The adjusted mean yield the average rate by which TBARS changes with time in each meat product, and the effect of the different tomato products on this rate is shown in Fig. 4 for increasing pressure. In accordance with Fig. 3, the pressure

treatment at 300 MPa was milder with respect to the induction of lipid oxidation, as the rate of development in TBARS during storage was not significantly different between the meat samples. However, at higher pressures the effect of addition of tomato products to the meat resulted in the following order to delay lipid oxidation: at 600 MPa: waste ~ final > finishing ~ in natura ~ control, and at 800 MPa: waste > final ~ finishing ~ in natura ~ control (where ~ denotes no mutual significant difference and > denotes significant better protection when comparing the treatments). In conclusion, only the tomato waste acted as an effective antioxidant in pressure-processed minced chicken breast meat during chill storage. The tomato waste, added in a 3 times higher amount than the others tomato products, results in a similar content of total phenolics and β-carotene, from 6 to 7 times less lycopene, and from 2 to 3 times more total flavonoids, rutin, quercetrin and naringenin (Fig. 1). The antioxidant protection seems accordingly mainly to be performed by the flavonoids present in the product.

The effect of increasing hydrostatic pressure used for meat processing on the subsequent progression of lipid oxidation during chill storage is clear, and the pressure treatment above 300 MPa accelerates lipid oxidation as may been seen in Fig. 4. Moreover, the rate of lipid oxidation appears not to increase linearly with pressure for chicken meat without tomato products added and with chicken meat with the in natura and finishing tomato products added. A similar non-linear relationship was previously demonstrated by Orlien et al. (2000), who established 500 MPa as a threshold pressure for whole chicken breast fillet with respect to lipid oxidation. Beltran et al. (2003) observed no effect on the oxidative stability of minced chicken breast subjected to pressure treatments for up to 500 MPa, supporting the suggestion of a threshold pressure. There is a tendency of a linear correlation between lipid oxidation development and applied pressure for the chicken meat added with the final and waste tomato products, a relationship also found for minced chicken breast (Mariutti et al., 2008) and turkey meat (Dissing et al., 1997). Other meat muscles seem to be susceptible to pressure-induced lipid oxidation, as pressure-treatment above 300 MPa of pork resulted in increased rates of lipid oxidation (Cheah and Ledward, 1996) and pressure-treatment at 400 MPa of cod increased the rate of oxidation (Angsupanich and Ledward, 1998).

α-Tocopherol and γ-tocopherol are the most important antioxidants naturally present in chicken meat, and the content of α- and γ-tocopherol were measured prior to storage and at the last day of storage in order to evaluate the interaction in the pressure-treated meat product between the tocopherols and the compounds present in the different tomato products. Raw chicken breast meat was found to have 3.97 ± 0.51 mg/kg meat and 0.35 ± 0.02 mg/kg meat of α- and γ-tocopherol, respectively. Slightly higher initial contents of the tocopherols were found for the products with addition of the different tomato products after pressure treatment, most likely due to tocopherols present in the tomato products. According to Capanoglu et al. (2008), tomato products contain between 24 and 39 mg/100 g of dry weight. A small decrease in the content of α-tocopherol occurred in the meat without and with addition of the different tomato samples during the storage time, while no significant changes in the γ-tocopherol contents were observed (results not shown). Apparently, no natural protection of α- and γ-tocopherol present in chicken meat by tomato antioxidants was observed. Similar results were found when rosemary was added in high pressure chicken meat (Bragagnolo et al., 2005).

High-pressure treatment of chicken meat was found to result in an altered appearance and all of the pressurized samples had higher L^* values indicating a more white color, an appearance similar to heat-treated chicken meat. In accordance, the pressurized meat products were found to have lower a^* values and b^* values and appeared less red. The color parameters changed from the raw meat to the pressurized meat, but were not affected by the pressure level or the storage time, irrespective of addition of tomato products (data not shown). It should, however, be noted that the tomato products were added in very small amounts in order not to contribute to the color of the meat. Similar meat discoloration resulting from a whitening effect and a loss of red color due to high pressure processing has previously been observed for chicken (Mariutti et al., 2008), beef (Carlez et al., 1995), and pork (Shigehisa et al., 1991), but the mechanism of pressure-induced color changes does not seem to have been established yet.

Two findings are of importance for strategies for production of high-pressure processed chicken meat using tomato fractions for antioxidant protection. The antioxidative effect is concluded not to be on the initial pressure-induced radical formation as lycopene would be an efficient singlet oxygen quencher and radical scavenger, but rather on the subsequent reactions leading to secondary lipid oxidation products for which the chain-breaking antioxidants like flavonoids become important. Moreover, the tomato waste seems to have a good application as antioxidant in high-pressure processed chicken as it has a high content of flavonoids.

Acknowledgements

The authors thank the Brazilian agencies CAPES, CNPq and FAPESP for the financial support and Predilecta Alimentos Ltda for the tomato samples.

References

Alves, A.B., Silva, M.G., Carvalho, P.R.N., Vissoto, L.C., 2010. Validação e estimativa da incerteza de método para análise de licopeno e beta-caroteno em polpa de tomate por cromatografia líquida de alta eficiência. Quimica Nova 33, 1962–1966.

Angsupanich, K., Ledward, D., 1998. High pressure treatment effects on cod (*Gadus morhua*) muscle. Food Chemistry 63, 39–50.

Beltran, E., Pla, R., Yuste, J., Mor-Mur, M., 2003. Lipid oxidation of pressurized and cooked chicken: role of sodium chloride and mechanical processing on TBARS and hexanal values. Meat Science 64, 19–25.

Beltran, E., Pla, R., Yuste, J., Mor-Mur, M., 2004. Use of antioxidants to minimize rancidity in pressurized and cooked chicken slurries. Meat Science 66, 719–725.

Bragagnolo, N., Danielsen, B., Skibsted, L.H., 2005. Effect of rosemary on lipid oxidation in pressure-processed, minced chicken breast during refrigerated storage and subsequent heat treatment. European Food Research and Technology 221, 610–615.

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. Innovative Food Science & Emerging Technologies 8, 24–29.

Capanoglu, E., Beekwilder, J., Boyacioglu, D., Hall, R., De Vos, R., 2008. Changes in antioxidant and metabolite profiles during

production of tomato paste. Journal of Agricultural & Food Chemistry 56, 964–973.
Carlez, A., Veciana-Nogues, M.T., Cheftel, J.C., 1995. Changes in Colour and Myoglobin of Minced Beef Meat Due to High Pressure Processing. Lebensmittel-Wissenschaft und-Technologie 28, 528–538.
Cheah, P.B., Ledward, D., 1996. High pressure effects on lipid oxidation in minced pork. Meat Science 43, 123–134.
Dissing, J., Bruun-Jensen, L., Skibsted, L.H., 1997. Effect of high-pressure treatment on lipid oxidation in turkey thigh muscle during chill storage. Zeitschrift für Lebensmitteluntersuchung und -Forschung A 205, 11–13.
Garcia, A.F., Butz, P., Tauscher, B., 2006. Effects of high-pressure processing on carotenoid extractability, antioxidant activity, glucose diffusion, and water binding of tomato puree (*Lycopersicon esculentum* Mill.). Journal of Food Science: Sensory and Nutritive Qualities of Food 66, 1033–1038.
Hsu, K-C., 2008. Evaluation of processing qualities of tomato juice induced by thermal and pressure processing. LWT-Food Science and Technology 41, 450–459.
Jensen, C., Guidera, J., Skovgaard, I.M., Staun, H., Skibsted, L.H., Jensen, S.K., Møller, A.J., Buckley, J., Bertelsen, G., 1997. Effects of dietary α-tocopheryl acetate supplementation on α-tocopheryl deposition in porcine m psoas major and m. longissimus dorsi and on drip loss, colour stability and oxidative stability of pork meat. Meat Science 45, 491–500.
Kim, D.O., Jeong, S.W., Lee, C.Y., 2003. Antioxidant capacity of phenolic phytochemicals from various cultivars of plums. Food Chemistry 81, 321–326.
Mariutti, L.R.B., Orlien, V., Bragagnolo, N., Skibsted, L.H., 2008. Effect of sage and garlic on lipid oxidation in high-pressure processed chicken meat. European Food Research and Technology 227, 337–344.
Mielche, M., Bertelsen, G., 1995. Evaluation of a mathematical model for prediction of lipid oxidation in heat-treated beef during chill storage. Zeitschrift für Lebensmitteluntersuchung und -Forschung 200, 415–419.
Mortensen, A., Skibsted, L.H., 1997. Real time detection of reactions between radicals of lycopene and tocopherol homologues. Free Radical Research 27, 229–234.
Nissen, L.R., Månsson, L., Bertelsen, G., Huynh-ba, T., Skibsted, L.H., 2000. Protection of dehydrated chicken meat by natural antioxidants as evaluated by electron spin resonance spectrometry. Journal of Agricultural & Food Chemistry 48, 5548–5556.
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. European Food Research and Technology 211, 99–104.
Roldán-Marín, E., Sánchez-Moreno, C., Lloría, R., Ancos, B., Cano, M.P., 2009. Onion high-pressure processing: flavonol content and antioxidant activity. LWT-Food Science and Technology 42, 835–841.
Sánchez-Moreno, C., Plaza, L., Ancos, B., Cano, M.P., 2006. Impact of high-pressure and traditional thermal processing of tomato purée on carotenoids, vitamin C, and antioxidant activity. Journal of the Science of Food and Agriculture 86, 171–179.
Shigehisa, T., Ohmori, T., Saito, A., Taji, S., Hayashi, R., 1991. Effects of high hydrostatic pressure on characteristics of pork slurries and inactivation of microorganisms associated with meat and meat products. International Journal of Food Microbiology 12, 207–216.
Singleton, V.L., Rossi Jr., J.A., 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American Journal of Enology and Viticulture 16, 144–158.
Sørensen, G., Jørgensen, S.S., 1996. A critical examination of some experimental variables in the 2-thiobarbituric acid (TBA) test for lipid oxidation in meat products. Zeitschrift für Lebensmitteluntersuchung und -Forschung 202, 205–210.
Wiggers, S.B., Kröger-Ohlsen, M.V., Skibsted, L.H., 2004. Lipid oxidation in high-pressure processed chicken breast during chill storage and subsequent heat treatment: effect of working pressure, packaging atmosphere and storage time. European Food Research and Technology 219, 167–170.
Zishen, J., Mengcheng, T., Jianming, W., 1999. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chemistry 64, 555–559.