Effect of dietary *Curcuma longa* L. powder on lipid oxidation of frozen pork

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SUMMARY

Introduction - Modification of the eating habits incremented the consumption of ready-to-eat, ready-to-cook and frozen foods. Meat can be store up as frozen for different time in relation to the animal species, to the cut or to a previous processing. Several researches were conducted to evaluate the use of antioxidant products as dietary supplementation in order to produce modification in chemical composition of meat and lead to a positive influence on the length of shelf life of the products.

Aim - The aim of this research was to study the effect of the supplementation with *Curcuma longa* L. powder in pig diet on the lipid oxidation after common frozen storage (-18 °C) of meat for long time (up to 135 days).

Materials and methods - Pigs were fed with two different diets, control and turmeric, for 30 days before been slaughtered. *Longissimus lumborum* chops were frozen at -18 °C and lipid oxidation was evaluated with the thiobarbituric acid reactive substances (TBARS) method after 45, 60, 75, 90, 105, 120 and 135 days of storage.

Results and discussion - The addition of the natural antioxidant did not modify the meat quality of the evaluated samples (P > 0.05). Nor the time of storage increased the TBARS value of the meat (P > 0.05). Our results are in accordance with other similar research studies on antioxidant dietary supplementations.

Conclusion - Pigs, as large animal, probably need a bigger amount of antioxidant supplementation in order to modified meat characteristics. Thus, the lack of difference in lipid oxidation of frozen meat showed by Control and Turmeric diets could be associated both to the small dose of dietary turmeric powder and to the length of the diet period.

KEY WORDS

Meat quality, frozen, TBARS, natural antioxidant, turmeric.

INTRODUCTION

Storage of foods changed throughout the history, following the technology and the life style of the different periods. Recently, modification of the eating habits, incremented the consumption of ready-to-eat, ready-to-cook and frozen foods¹.

Several studies and reports refer the habits of consumers to buy frozen meat products, or buy raw meat and then froze it as block or slices, as a common procedure to reduce waste of time².

The FAO itself suggests freezing meat "when the preservation period is longer than that acceptable for chilled meat, freezing must be used to minimize any physical, biochemical and microbiological changes affecting quality in storage"³. The United States Department of Agriculture (USDA) by the department of Food Safety and Inspection Service (FSIS) suggest storing for not more than 4-12 months the meat as uncooked steaks or chops in order to amend not excessively the quality⁴. Meat can be store up as frozen for different time in relation to the animal species, to the cut or to a previous processing. Modification in chemical composition of meat before the storage can also have their influences on the time of storage. Several researches were conducted in different species, to evaluate the use of antioxidant ingredient as dietary supplementation during rearing period^{5,6,7}, and other were con-

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ducted to verify the lipid peroxidation and the antioxidant activity on meat products added with different natural antioxidant, stored for different periods or processed^{8,9,10}.

Curcuma longa L. (turmeric) is widely studied for its antioxidant properties both *in vivo* and *in vitro*. Jain et al.¹¹ reported the significant medical potential of turmeric as anti-inflammatory, anti-infectious and anti-tumour agent. One of the main components of turmeric are curcuminoids (curcumin, demethoxycurcumin and bisdemethoxycurcumin) with high antioxidant activities. Curcuminoids are in relation with the inhibition or delay of lipid peroxidation *in vitro* with comparable activity to vitamin C or synthetic antioxidants (such as BHT, butylated hydroxyltoluene)¹². Moreover, turmeric is rich in both hydrophobic and hydrophilic compounds such as monoterpenes, sesquiterpenes, phenols and alcohols. These molecules represent an important source of natural antioxidant and could play an important role as modulator of the intestinal microbiota¹³.

Lipid peroxidation is one of the most important parameter of quality assessment in meat and meat products. The natural enhancement of lipid peroxidation in meat produces off odour, off flavour and the decrement of nutritional value. Thiobarbituric acid reactive substances (TBARS) method is widely used to determine the lipid peroxidation *status* of a food matrix. In the present study supplementation of *Curcuma longa*

powder in pig diet have been evaluated in order to quantify the lipid peroxidation (expressed as TBARS value) after common frozen storage (-18 °C) of pig's meat for long time (up to 135 days).

MATERIAL AND METHODS

This study was carried out on meat derived from 18 Cinta Senese pigs. Pigs were reared outdoors in pasture for 15 months (live weight 122 ± 2 kg) than pigs were randomly divided in six groups and assigned to two different dietary treatments (D, three groups for diet). At three groups no supplementation was added (Control groups) as reported in Mancini et al.¹⁴ and were fed with a basal diet of 2 kg of feed (barley 50%, wheat 25%, beans 15% and oats 10%). At three groups were added daily fresh 4.5 g/pig of *Curcuma longa* powder (protein 12.2%, fat 3.4%, ash 5.8%, and moisture 9.4%; Turmeric groups).

After 30 days of the dietary treatments all the animals were electrically stunned and slaughtered at a local commercial slaughterhouse. After 24h at 4°C the right *Longissimus lumborum* muscles were sectioned and transported to the laboratory.

From each pig seven 25 mm thick chops were cut perpendicular to the fibers direction. All meat samples were frozen individually at -18°C in polyethylene bags under vacuum. One chop from each pig was analysed for quantified lipid peroxidation at one selected day of storage time (ST: 45, 60, 75, 90, 105, 120 and 135 days).

At the assigned storage time eighteen samples (9 for each diet treatment) were thawed (at $4 \pm 1^{\circ}$ C for approximately 18 h). Thiobarbituric acid reactive substances (TBARS) were assessed in accord to Mancini et al.¹⁵.

Five g of sample was homogenized for 45 s at 9000 rpm (Polytron PT 3000, Kinematica AG, Eschbach, Deutschland) with 10 mL of 7.5% trichloroacetic acid (TCA) and 0.1% diethylenetriaminepentaacetic acid (DTPA) in distilled water (final concentration).

The homogenized sample was centrifuged (10,000 rpm for 10 min) (4235A CWS, ALC International, Milan, Italy) and filtered through Whatman number 1 filter paper, and 5 mL of the filtrate was mixed with 2.5 mL of 2-thiobarbituric acid (TBA) solution (0.288% in distilled water) in capped test tubes. The tubes were vortexed and placed in a water bath at 95 °C for 45 min, then cooled under tap water.

The absorbance was determined at 532 nm (V-530 Jasco International, Milan, Italy) against a blank containing trichloroacetic acid (TCA) and diethylenetriaminepentaacetic acid (DTPA) solution instead of a sample extract. TEP (1,1,3,3tetraethoxypropane; 0-15 μ M, final concentrations) was used to obtain a calibration curve and the results were expressed as μ g of malondialdehyde (MDA) per kilogram of sample. Determinations were performed in triplicate on each chop and the mean value was calculated.

Data were analysed with the following linear model:

$$Y_{ijkz} = \mu + \alpha_i + \beta_j + \gamma_k + \alpha\beta_{ij} + e_{ijkz}$$

Where:

 Y_{ijkz} = dependent variable of the z^{th} observation;

 μ = overall mean;

 α_i = effect of the diet, D (*i* = Control, Turmeric);

 β_j = effect of the storage time, ST (*j* = 45, 60, 75, 90, 105, 120, 135 days);

 γ_k = effect of the block B (*k* = 1, 2, 3, 4, 5, 6);

 $\alpha \beta_{ij}$ = effect of the interaction between D and ST (D x ST); e_{ijkz} = random error.

After a first statistical analysis B was removed from the final model because no block significant difference was found. The lipid peroxidation was analyzed by two-ways ANOVA method using R free statistical software.

RESULTS AND DISCUSSION

The effects of diet (D) and storage time (ST) as well as their interaction (D x ST) on lipid peroxidation are shown in Fig. 1. The TBARS did not differ among the experimental factors considered (P>0.05 for both D and ST), even if control meat samples showed slightly higher TBARS values.

During storage time TBARS reported a not statistically significant trend with slightly lower values after 90 and 105 days of storage, likely due to slightly lower values of TBARS of turmeric diet samples (P>0.05) (Fig. 1).

The TBARS values of frozen samples were ranged between 29.45 μ g MDA on kg of sample (for turmeric diet samples at 60 days) and 31.77 μ g MDA on kg of sample (for control di-



Figure 1

Thiobarbituric acid reactive substances (TBARS) values expressed as µg malondialdehyde (MDA) on kg of sample of raw frozen meat samples as function of the interaction of diet x storage time (D x ST).

Control diet samples (basal diet);

E: Turmeric diet samples (basal diet supplemented with 4.5 g/pig/day of turmeric powder). et samples at 120 days). In particular, the TBARS of raw frozen meat samples derived from pigs fed with turmeric remained at slightly lower levels than those derived from pigs fed with control diet with the exception of 75 days storage time, anyway these trend was not supported by statistical analysis (P>0.05).

The frozen samples showed a different trend than that observed in raw samples, stored at 4°C for 7 days, which showed significant differences in lipid peroxidation within diets and storage time¹⁴. Botsoglou et al.⁵ reported similar results on lipid peroxidation of meat of chicken fed with a natural antioxidant, oregano essential oil, and α -tocopherol acetate during a long frozen storage at -20°C (9 months).

Our results are consistent with those of Sayago-Ayedi et al. in 2009¹⁶ who reported how dietary grape pomace concentrate (GPC) in chicken at level of 30% and 60% did not differ in TBARS values from control breast patties at day 0; after 6 months of frozen storage (-20°C) lipid peroxidation was increased for all the treatments. In this research was reported a lack of difference in lipid peroxidation between control and 30 GPC after 6 months of frozen storage, only the highest concentration of GPC in the diet delay the lipid peroxidation. Other research showed efficient effects of natural antioxidant on lipid peroxidation of raw samples in chicken meats during frozen conservation after feeding tea catechins; natural antioxidants inhibit lipid peroxidation in chicken breast and thigh frozen at -20°C for 9-12 months¹⁷.

Larger animals, as well as pigs, probably need a bigger amount of antioxidant supplementation in order to modified meat characteristics. Moreover, the lipid composition of the meat can induce different responses to the antioxidant provided. Antioxidant capacity expressed as ABTS, DPPH and FRAP values of the raw meat samples did not differ between Control and Turmeric diet, as reported by Mancini et al.¹⁴, and it could have influenced the effectiveness of Turmeric diet to inhibit the lipid peroxidation during the frozen storage period. Thus, the lack of difference in lipid oxidation of frozen meat showed by Control and Turmeric diets could be associated both to the small dose of dietary turmeric powder and to the length of the diet period.

Hanczakowska et al.¹⁸ reported that dietary natural antioxidant products, derived from herbal extracts, reduced TBARS in long term frozen period (5 months) even if no differences were detected after 24 h of cooling. Similarly, dietary supplementation with dried fruit and vegetable pomaces reduced TBARS in pork after 3 months of frozen storage¹⁹.

Even a well known antioxidant compound as Vitamin E reported different response in lipid oxidation as dietary supplement as function of the dose and the length of the dietary period.

Coronado et al. in 2002^{20} reported how both dietary supplementation of Vitamin E (200 mg/kg feed for 10 weeks) in pigs and addition of rosemary extracts in wiener sausages lacks to affected the TBARS values after a frozen storage at -20°C for 10 months. Popova et al. in 2014⁶ reported that dietary supplementation of vitamin E (400 mg/kg feed for 10 weeks) in swine modified the lipid peroxidation of the muscles after 48 h and 6 days of refrigeration at 4°C and after 90 days of frozen period. Meat samples derived from the pig that received vitamin E showed in all the tested retail times lower TBARS values that the samples of the control group.

CONCLUSION

In conclusion, the results of the present study show that, with the used doses, dietary *Curcuma longa* powder supplementation is not able to reduce significantly the lipid oxidation in meat submitted to long frozen storage (-18°C up to 135 days).

Higher doses of turmeric powder supplementation are suggested in order to investigate in-depth its effect on meat quality characteristics in long frozen storage.

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