INJECTION OF PORK LOINS WITH THE LONG CHAIN FATTY ACID DOCOSAHEXAENOIC ACID (DHA) AND ITS EFFECT ON FLAVOUR.

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Abstract - Pork is traditionally low in docosahexanoic acid (DHA, C22:6n-3) and deficient in omega-3 fats for a balanced human diet. Treatments of a mixed brine control (CON), 3.1% sunflower oil in mixed brine (SF) and a 3.1% DHA as a oil triglycerides in mixed brine (DHA) were injected into pork loins at 10 mL/100 g and grilled at 205°C. After cooking, the CON and SF pork loins contained 0.03 to 0.05 mg DHA per gram of pork and the DHA injected loins contained approximately 1.16 mg DHA per gram. The appearance, odor, oxidation rates and sensory taste, as judged by a trained panel, determined the DHA injected meat to be, 'slightly desirable' and gave lower `off odour' scores relative to the CON and SF injected pork. Pork can be fortified with DHA oil, which would meet half the recommended omega 3 fatty acid requirements and would be acceptable in taste.

Keywords - Docosahexaenoic acid, Pork, Injection marinade, Sensory characteristics

I. INTRODUCTION

Pork is viewed as a lean healthy food, providing good nutrition; however, there are concerns about the quantity and types of fat it possesses. According to the USDA, a typical pork chop contains 11.3 g of fat per 100 g of which, 1.3 g is polyunsaturated fat, and essentially no omega-3 fats [6]. Human adults are recommended to consume at least 1 g per day of omega-3 fat for proper cardiovascular health. The long chain omega-3 fatty acid, docosahexaenoic acid (C22:6n-3), is particularly important, since it comprises ~14% of the cerebral corte. Pork can be selectively enriched with DHA by feeding microalgae biomass Schizochytricium [13]. The option of directly injecting the DHA into the meat as a brine marinade was explored.

Injecting water for moisture into pork has been in practice since 1960 [3]. The addition of a polyphosphate to a brine mixture further improves the juiciness, tenderness and flavour after cooking [7]; however, some discoloration has been noted. In addition to brine, injection of fats and oils may improve the eating experience of pork. In North America, lean pork loins are averaging less than 2% intramuscular fat (IMF), the minimal IMF for consumer acceptance is >3% [9]. The IMF adds flavour and juiciness and has a minor improvement on tenderness. Beef injections with conjugated linoleic acid has recently been done to improve the nutrition but also to improve the eating quality experience of beef [2]. These studies was done to improve the nutritional profile of pork by injecting lean pork loins with DHA oil and to assess consumer perceptions of eating quality and to examine if any off flavours would be generated by the DHA oil.

II. MATERIALS AND METHODS.

Three treatments were randomly allocated to the boneless loins (n=24). The treatments were an injection of 10 ml per 100 g loin (longissimus dorsi muscle) of mixed brine solution (CON) containing phosphate, sodium chloride, a 3.1% sunflower oil in the mixed brine solution (SF) and a 3.1% DHA oil in mixed brine solution (DHA). The mixed brine consisted of, 4.8% sodium tripolyphosphate Na₅P₃O₁₀ (BCCHEM, PQ, Canada), 4.8% sodium chloride, 0.01% αtocopherol, and 0.15% precept 8140 powdered soy lecithin in distilled water. The SF oil consisted of the control brine mixed with 3.1% of mid oleic grade sunflower oil (Compliments, ON, Canada). The DHA oil consisted of the control brine mixed with 3.1% of DHA-S oil (Martek Bioscience Corp, Boulder, CO, USA). The brine mixtures were injected using 4 mm needles spaced 2.8 cm in an Inject Star BI-72

unit (J Redmond & Sons, Northampton, UK), set at 2 bar, 56 strokes/min. The day 1 raw chops were evaluated by trained panellists for visual colour, stripping caused by the injections and odours and measured for color and thiobarbituric acid (TBARS) and again after 3 days under refrigeration at 2°C. Fatty acid methyl esters (FAME) extracts were isolated from the DHA and SF oils. FAME were also isolated from 12 of the thawed, CON, SF and DHA raw and 12 of the equivalent cooked injected chops and were prepared according to the method of (Sukhija and Palmquist, 1988). Chromatograms were integrated using Varian Star Chromatography Workstation software. Peaks were identified using a GC reference standard GLC463 from Nu-Check-Prep, Elysian, MN, USA). The iodine value of the fatty acids was calculated. The colour of each loin treatment section was measured using a Minolta CM2002 color meter (Minolta Canada Inc., ON, Canada). (Konica Minolta, Ramsay, NJ, USA). TBARS values were determined relative to a standard curve of malonaldehyde generated with 1 g/L of tetraethoxypropane and 20 mM to 90 mM TBA solution [14]. Panellist (n=8) were selected and trained according to the American Meat Science Associations guidelines [1]. The panellist were asked evaluate the visual display of the 0 d and 3 d raw loin chops and give rating based on a 8scale. Odour rating was point hedonic completed using a 4-point descriptive scale. The injected loin chops were sliced into 1 inch chops and then cooked on a preheated Garland electric grill ED-30B at 205°C. The panellist were asked to rate the samples on 9-point descriptive scale. Statistical analysis on the lipid profiles were analyzed using the MIXED procedure and significance was determined by the PDIFF procedure to identify differences between the groups CON, SF, and DHA or by raw and cooked treatment effect [15]. The statistical model included a treatment by the number of days, interaction. An ordinate scale was used for the panellist evaluations of the retail appearance using Friedman test and the nominal scale was used for the biochemical measurement values using Tukey's HSD test.

Sunflower (SF) and DHA treatments.					
FAME	Cooked	Pork			
mg/ g	CON	SF	DHA	SEM	Prob.
wet tissue C14:0	0.55A	1.44C	1.68C	0.130	0.004
C16:0	10.54A	26.65C	24.64BC	1.524	0.001
C16:1cis9	1.11A	2.60C	2.70C	0.164	0.002
C18:0	5.74A	14.78C	12.09B	0.833	0.001
C18:1cis9	15.29A	40.42C	33.79C	2.002	0.001
C18:1cis11	2.13A	5.61B	4.95B	0.335	0.177
C18:2n-6	3.05A	6.64C	5.14B	0.414	0.001
C18:3n-3	0.32	0.87	0.76	0.089	0.296
C18:3n-6	0.09	0.28	0.20	0.021	0.005
C20:1cis11	0.37A	1.16C	0.83B	0.075	0.004
C20:2n-6	0.07A	0.21C	0.15B	0.016	0.023
C20:3n-6	0.05	0.10	0.10	0.009	0.258
C20:3n-3	0.03	0.07	0.06	0.008	0.102
C20:4n-6	0.33A	0.45B	0.37A	0.022	0.001
C20:5n-3	0.06	0.07	0.08	0.005	0.002
C22:0	0.01	0.02	0.02	0.003	0.073
C22:5n-6	0.01A	0.00A	0.54B	0.025	0.108
C22:5n-3	0.12	0.21	0.18	0.016	0.002
C22:6n-3	0.03A	0.05A	1.46B	0.068	0.075
SFA ^x	16.93A	43.17B	38.63B	2.510	0.001
MUFA ^y	18.90A	49.78C	42.28BC	2.576	0.001
PUFA ^z	4.06A	8.67B	8.83B	0.673	0.000
Total	39.88A	101.62C	89.73B	1.920	0.001
FAME Iodine Index†	26.32A	64.48B	60.95B	3.184	0.001

Table 1 Fatty acid methyl esters profile of the Cooked injected pork loins between CON, Sunflower (SE) and DHA treatments

ABC - Means within column, with unique letter, differ significantly (P > 0.05). SEM; standard error of means within row.

^xSFA; saturated fatty acids, with no double bonds.

^yMUFA; monounsaturated fatty acids, with one double bond.

^zPUFA; polyunsaturated fatty acids, with two or more double bonds.

*Estimated iodine index calculated by the % of fatty acid with the sample multiplied by the iodine value of the fatty acid.

III. RESULTS AND DISCUSSION

The injected loin treatments were primarily performed to determine if the DHA oil could be added a concentration of 1 mg per gram of fresh pork, without adversely affecting aroma or taste. Regular pork loins from pigs fed a standard finisher diet of, corn, barley, peas, and canola, would have ~0.5 mg of omega-3 FAME /g of meat and only ~0.02 mg of DHA FAME /g of meat [6]. Injection of the DHA brine mixture at 10 mL per 100 g into the boneless meat. increased the DHA (C22:6n-3) content 50-fold, to an estimated concentration of 1.05 mg/g of pork. The actual concentration in the loin chops was 1.16 mg/ g of raw pork (Table 2). The retention of DHA was higher after cooking at 1.46 mg/g of cooked pork, due to water loss by cooking (Table 1). The average cooking loss for all three treatments was $21.5 \pm 3.04\%$. Conservatively, this would adjust the estimated level of DHA to approximately 0.82 mg/g of pork, if the oil was retained evenly but usually fatty acid content is increased by cooking [2]. The amount of 18:1cis9 and 18:2n-6 was also significantly increased in the SF and DHA treatments but the final concentration of 18:1cis9 and 18:2n-6 was increased less than 2fold in the actual raw and cooked pork. Sov lecithin was added to the mixture because it was needed to assist the emulsion of the sunflower oil and DHA oil. In the CON mixture, the addition of the soy lecithin will impart a slightly nutty aroma [17]. The vitamin E (α -tocopherol) was added to help maintain oxidative stability of the oil injection mixture and was considered as odourless. The addition of vitamin E to the injected chops was expected to help prevent rancid odours and flavours [5]. The brine's main ingredients were 4.8% sodium tripolyphosphate and 4.8% sodium chloride and were also determined to be odourless by the sensory panel. Injection of a brine mixture increases tenderness and juiciness and might add some saltiness to the flavour while reducing the intensity of the pork flavour [8]. The panellists also did not detect any difference in the retail display, marbling, or striping between the treatment groups. However, in the injected chops in the display case for 3d, the panellists did give poorer scores for overall retail display and detected some color striping in the CON and treated chops. The objective colour score measured by the Minolta color meter showed a difference between the 1d and 3d chops.

The degree of oxidation, as indicated by the amount of malonaldehyde generated by lipid peroxidation, was measured using the TBARS assay. The injected loin chops and the purge juice from the meat samples were collected from the 1 day and 3 day retail display packages. On day 1, the degree of oxidation was negligible according to the assay. On day 3, the amount of oxidation in the purge but not the meat, was significantly higher compared to day 1 but not between treatment groups. Meat purge represents the free flowing juices around the meat and may have a greater chance of interacting with the atmospheric oxidation. The DHA oil had nearly twice the estimated iodine value at 271.36 than the sunflower oil at 110.43 (Table 1) and therefore the potential for lipid oxidation would be expected to be greater [10]. The sensory panellists judged the DHA and SF injected raw pork to be both worse for odour unacceptability after 3 d, than the CON pork.

The odour and sensory evaluations were made with a 10 member trained taste panel. There was a noticeable drop in the 'unacceptable odours' score by day 3 but this was still within the partially acceptable to neutral range and consistent between all treatments. There was no difference between the treatments and the scores were very low and unchanged for `off' odours in the day 1 and day 3 chops. Chops were cooked, 24 h after injection. The amount of cooking loss (%) was not significantly different between the CON (22.2 $\pm 2.8\%$), the SF (19.9 $\pm 3.6\%$) or the DHA (22.4 $\pm 2.2\%$) treatments. The injected cooked chops rated highly for scores of, juiciness, tenderness, and salt intensity. Juiciness was scored the highest in the DHA injected chops. The brine injected meat has only a minimal increase on saltiness scores and a less intense, pork flavour. It has been speculated that the flavour of brine injection, dilutes the carbohydrates, proteins and lipids and washes away the Maillard reactions complexes, which

give meat its' roasted flavours [4]. If a panellist did mark the injected chops for `off flavours', they scored the sample as very low and gave a description as `stale' or `piggy' and surprisingly, the off flavours score were higher in the untreated CON and SF injected cooked pork than the DHA injected cooked pork (table 5). It has been noted that DHA triacylglycerol can impart umami and flavour and supress bitterness in certain taste panels [11].

IV. CONCLUSIONS

The injection of pork loins with 3.1% DHA in a tripolyphosphate brine mixture appears to be well accepted by trained taste panellist. The addition of DHA oil added to the nutritional value of the pork will help in reducing plasma triglycerides of consumers [12]. DHA content was improved approximately 50- fold to 1.16 mg DHA per gram of raw pork, which converts to 116 mg of DHA in a typical 100 g serving of pork. The DHA content was improved by cooking on a grill to 146 mg of DHA / 100 g of pork. This would meet over half the recommended daily requirements for DHA omega-3 fatty acid in healthy human diets [16]. The trained taste panel did score the cooked DHA injected pork better at surviving against off flavours, than CON and SF pork.

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- 1. AMSA. (1995). Research guidelines for cookery, sensory evaluation and instrumental measurements of fresh Meat. Chicago, IL, USA.: American Meat Science Association.
- Baublits RT, Pohlman FW, Brown AH, Jr., Johnson ZB, Proctor A, Sawyer J, Dias-Morse P & Galloway DL. (2007). Injection of conjugated linoleic acid into beef strip loins. Meat Sci 75(1):84-93.
- 3. Bendall JR. (1954). The swelling effect of polyphosphates on lean meat. Journal of the Science of Food and Agriculture. 5:468-475.
- Blake A. (1975). Flavour of meat with respect to the Maillard reaction. Afinidad 32(328):615-618
- 5. Cardenia V, Rodriguez-Estrada MT, Cumella F, Sardi L, Della Casa G & Lercker G. (2011).

Oxidative stability of pork meat lipids as related to high-oleic sunflower oil and vitamin E diet supplementation and storage conditions. Meat Sci 88(2):271-279.

- Daniel CR, Cross AJ, Koebnick C & Sinha R. (2011). Trends in meat consumption in the USA. Public Health Nutr 14(4):575-583.
- Detienne NA, Reynolds AE & Wicker L. (2003). Phosphate marination of pork loins at high and low injection pressures. Journal of Food Quality 26(1):1-14.
- Detienne NA & Wicker L. (1999). Sodium chloride and tripolyphosphate effects on physical and quality characteristics of injected pork loins. Journal of food science. Nov/Dec 64(6):1042-1047.
- Eikelenboom G, Hoving-Bolink AH & van der Wal PG. (1996). The eating quality of pork. 2. The influence of intramuscular fat. Fleischwirtschaft 76(5):517-518, 559-560.
- Knothe G. (2002). Structure indices in FA chemistry. How relevant is the iodine value. Journal of the American Oil Chemists' Society. Sept 79(9):847-854.
- Koriyama TKTWKAH. (2002). Effects of docosahexaenoic acid content in triacylglycerol on human taste perception. Journal of Food Science 67(6):2352-2356.
- Meadus WJ, Duff P, Rolland D, Aalhus JL, Uttaro B & Dugan MER. (2011). Feeding docosahexaenoic acid to pigs reduces blood triglycerides and induces gene expression for fat oxidation. Canadian Journal of Animal Science 91(4):601-612.
- Meadus WJ, Duff P, Uttaro B, Aalhus JL, Rolland DC, Gibson LL & Dugan ME. (2010). Production of docosahexaenoic acid (DHA) enriched bacon. Journal of Agricultural and Food Chemistry 58(1):465-472.
- Nielsen JH, Sorensen B, Skibsted LH & Bertelsen G. (1997). Oxidation in pre-cooked minced pork as influenced by chill storage of raw muscle. Meat Sci 46(2):191-197.
- SAS. (2003). SAS user's guide: Stastics. SAS for windows, version 9.1.: SAS Institute Inc., Cary, NC, USA.
- Simopoulos AP, Leaf A & Salem N, Jr. (1999). Essentiality of and recommended dietary intakes for omega-6 and omega-3 fatty acids. Ann Nutr Metab 43(2):127-130.
- 17. Stephan A & Steinhart H. (1999). Identification of character impact odorants of different soybean lecithins. Journal of Agricultural and Food Chemistry 47(7):2854-2859.

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