

Title	Enhancing the health-status of processed meats through ingredient manipulation and its effects on sensory and physiochemical product attributes
Authors	Tobin, Brian David
Publication date	2013
Original Citation	Tobin, Brian D., 2013. Enhancing the health-status of processed meats through ingredient manipulation and its effects on sensory and physiochemical product attributes. PhD Thesis, University College Cork.
Type of publication	Doctoral thesis
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Download date	2025-05-07 03:48:59
Item downloaded from	https://hdl.handle.net/10468/1017



University College Cork, Ireland Coláiste na hOllscoile Corcaigh

THE NATIONAL UNIVERSITY OF IRELAND UNIVERSITY COLLEGE CORK SCHOOL OF FOOD AND NUTRITIONAL SCIENCES



Enhancing the Health-Status of Processed Meats through Ingredient Manipulation and its Effects on Sensory and Physiochemical Product Attributes

A Thesis Submitted in the Fulfilment of the Requirements for the Degree of Doctor of Philosophy

Presented by

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Acknowledgements

This dissertation was funded by the Food Industry Research Measure as part of the Meat Matrix project. I would like to extend my gratitude towards FIRM for giving me the opportunity to work on this project and for funding my research for the past 4 years. I would also like to thank the Food and Nutritional Sciences Department at University College Cork for providing an excellent work environment and for looking after me while I completed my Ph.D.

I would like to take this opportunity to also thank the following people:

Dr. Joseph Kerry from the Food Packaging Group in UCC, for having the faith in my ability and character as an undergraduate which lead to my chance at completing a Ph.D under your supervision. It was an opportunity of a lifetime and I'm thankful that I had your supervision all along the way.

Dr Maurice O'Sullivan from the Food Packaging Group in UCC, without your supervision I would not have made it this far. You helped guide my thesis and myself through the past 4 years. You were patient, supportive and always had a great idea or two. For all your help you have my sincerest gratitude and I hope you enjoyed working with me as I you.

To the technicians in the Food Science Building in UCC, Thank you so much for all the help and training you gave me over the course of my time in UCC. You were all always available to help anytime and I have no doubt that with out you my thesis would not be where it is today. In particular I would like to thank Dr. Michael O' Grady, Dr. Yvonne O'Callaghan, James McNamara, Donal Humphreys, Diarmuid O' Dwyer and Maurice Conway. A special thanks to Eddie Beatty who thought me everything I know about working with many, many different pieces of processing and analytical equipment from day one of my Ph.D.

Kevin Murphy, Karen O'Callaghan, Andreas Hempel, Courtney Refakes, Nur Hanani, Natasha Moroney and everyone else that has ever worked with me in office 046, thank you for being my friends and for being some of the best people that I've ever known and it has been a pleasure to work you all.

I wish to thank my friends that I grew up with and made along way in my life so far. You all mean so much to me and even though you didn't directly help with my thesis you all helped me in my life and without your friendship I could never have gotten so far.

Finally I want to thank my family for not only raising me but also for always being there for me through all the ups and downs of a Ph.D. My deepest thanks go to my parents for all their love and support throughout my life.

Abstract

An important component of this Ph.D. thesis was to determine the European consumers' views on processed meats and bioactive compounds. Thus a survey gathered information form over 500 respondents and explored their perceptions on the healthiness and purchase-ability for both traditional and functional processed meats. This study found that the consumer was distrustful towards processed meat, especially high salt and fat content. Consumers were found to be very pro-bioactive compounds in yogurt style products but unsure of their feelings on the idea of them in meat based products, which is likely due to the lack of familiarity to these products.

The work in this thesis also centred on the applied acceptable reduction of salt and fat in terms of consumer sensory analysis. The products chosen ranged in the degree of comminution, from a coarse beef patty to a more fine emulsion style breakfast sausage and frankfurter. A full factorial design was implemented which saw the production of twenty beef patties with varying concentrations of fat (30%, 40%, 50%, 60% w/w) and salt (0.5%, 0.75%, 1.0%, 1.25%, 1.5% w/w). Twenty eight sausage were also produced with varying concentrations of fat (22.5%, 27.5%, 32.5%, 37.5% w/w) and salt (0.8%, 1%, 1.2%, 1.4%, 1.6%, 2%, 2.4% w/w). Finally, twenty different frankfurters formulations were produced with varying concentrations of fat (10%, 15%, 20%, 25% w/w) and salt (1%, 1.5%, 2%, 2.5%, 3% w/w). From these products it was found that the most consumer acceptable beef patty was that containing 40% fat with a salt level of 1%. This is a 20% decrease in fat and a 50% decrease in salt levels when compared to commercial patty available in Ireland and the UK. For sausages, salt reduced products were rated by the consumers as paler in colour, more tender and with greater meat flavour than higher salt containing products. The sausages containing 1.4 % and 1.0 % salt were significantly (P<0.01)

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found to be more acceptable to consumers than other salt levels. Frankfurter salt levels below 1.5% were shown to have a negative effect on consumer acceptability, with 2.5% salt concentration being the most accepted (P<0.001) by consumers. Samples containing less fat and salt were found to be tougher, less juicy and had greater cooking losses. Thus salt perception is very important for consumer acceptability, but fat levels can be potentially reduced without significantly affecting overall acceptability. Overall it can be summarised that the consumer acceptability of salt and fat reduced processed meats depends very much on the product and generalisations cannot be assumed.

The study of bio-actives in processed meat products found that the reduced salt/fat patties fortified with CoQ10 were rated as more acceptable than commercially available products for beef patties. The reduced fat and salt, as well as the CoQ10 fortified, sausages were found to compare quite well to their commercial counterparts for overall acceptability, whereas commercial frankfurters were found to be the more favoured in comparison to reduced fat and CoQ10 fortified Frankfurters.

Keywords: Processed Meat, Beef Patty, Frankfurter, Sausage, Functional Food, Bioactive, Co-Enzyme Q10, Sensory Evaluation, Physiochemical Evaluation, Consumer Survey.

Publication List

Published:

- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). Effect of varying salt and fat levels on the sensory quality of beef patties. *Meat Science*, 91, 460-465.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Effect of varying salt and fat levels on the sensory and physiochemical quality of frankfurters. *Meat Science*, 92, 659-666.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). The impact of salt and fat level variation on the physiochemical properties and sensory quality of pork breakfast sausages. *Meat Science*, 93, 145-152.)

Submitted:

- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). European Consumer Attitudes on the Associated Health Benefits of Neutraceutical-Containing Processed Meats using Co-Enzyme Q10 as a Sample Functional Ingredient Submitted to *Meat Science* on Sep 24, 2012; Status: Under Review.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Consumer Evaluation of the Commercial Viability of Reduced Salt and Fat Beef Patties and Patties Submitted to *Meat Science* on Dec 07, 2012; Status: Under Review.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Consumer Evaluation of Reduced Salt and Fat Sausages and Frankfurters and Variants Fortified with Co-Enzyme Q10. Submitted to *LWT* on Dec 07, 2012; Status: Under Review.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Effect of cooking and *in vitro* digestion on the stability of Co-Enzyme Q10 in processed meat products. Submitted to *Food Chemistry* on Dec 14, 2012; Status: With Editor.

Abstracts:

- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). Effect of varying salt and fat levels on the sensory and physiochemical quality of frankfurters. *Abstracts from the5th European Sensory Conference, Eurosense, Berne, Switzerland, 9th - 12th September 2012, P1.4.*
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). The impact of salt and fat level variation on the physiochemical properties and sensory quality of pork breakfast sausages. Abstracts from the5th European Sensory Conference, Eurosense, Berne, Switzerland, 9th - 12th September 2012, P1.3.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). Sensory quality of salt and fat reduced frankfurters *Abstracts from the 41st Annual UCC Food Research Conference, Cork, Ireland, 26th 27th April 2012, P13.*

- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). The sensory quality of pork breakfast sausages with varying levels of salt and fat. Abstracts from the 41st Annual UCC Food Research Conference, Cork, Ireland, 26th – 27th April 2012, P14.
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2011). Investigation of the comminuted meat matrix system for the development of low fat and low salt processed meat products. *Abstracts from the 40th Annual UCC Food Research Conference, Cork, Ireland, 31st March 1st April 2011, A38.*
- Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2010). Sensory consumer evaluation of reduced fat and salt beef burgers. *Abstracts from the4th European Sensory Conference, Eurosense, Vitoria Gasteiz, Spain, 5th - 8th September 2010, P1.038.*

Thesis Overview Schematic

Enhancing the Health-Status of Processed Meats through Ingredient Manipulation and its Effects on Sensory and Physiochemical Product Attributes



Chapter 1. Introduction

Processed meat products are an important category of food throughout the globe. Not only are they readily enjoyed by consumers worldwide, but they carry out vital requirements for the meat industry. The production of processed meat aims to take less commercial and usable forms of raw muscle which in some cases would otherwise be treated as waste and covert them into food products that are both economical and palatable. Processed meats are convenient and supply variety to the meat portion of the diet, these are the fundamental qualities of processed meats.

Many processed meat products have existed for millennia (Pearson & Gillett, 1996), their origin date is not known but likely around the time humans first learned of the preservative capabilities of salt. Initially meats were processed in an attempt to extend shelf life by the inhabitation of microbial decomposition. This could be done by salt addition, drying meat in the sun in warmer parts of the world or by burying meat in the snow in colder climates. As time progressed so did the technology; new chemicals were used such as nitrites, lactic acid bacteria were used to ferment the meat and new processes such as canning were invented. In more recent history, freeze drying, high pressure treatment and irradiation were developed and applied to meats. Over the course of human history many traditional meat products have been developed throughout the world and in some areas hold cultural importance.

Nowadays, meat and processed meat products have come under substantial scrutiny due to public health scares particularly; the emergence of bovine spongiform encephalopathy (BSE) in the late 1980's, the foot and mouth epidemic in the 2000's, excessive use of antibiotics and the illegal use of anabolic steroids to promote growth coupled with the reported association between a high level of meat consumption and ill health. In conjunction with the aforementioned health concerns, animal welfare issues and concerns over the impact of meat production on the environment have all

economically impacted the meat industry and formed negative attitudes toward meat in the consumers mind (Brewer & Rojas, 2008; Coffey, Mintert, Fox, Schroeder & Valentin, 2005; Garnier, Klont & Plastow, 2003; Thompson, Muriel, Russell, Osborne, Bromley, Rowland, Creigh-Tyte & Brown, 2002).

With an ever increasing number of people developing non-communicable diseases, such as heart disease, stroke, cancer, chronic respiratory diseases and diabetes in the world consumers are starting to pay greater attention to their diets contribution in our overall health (Fonseca & Salay, 2008; Angulo & Gil, 2007; Mollet & Rowland, 2002).

Processed foods have been stated as the main source of sodium in the human diet (Appel & Anderson, 2010). Reports linking a high intake of sodium to the incidence of hypertension (Law, Frost, & Wald, 1991a, 1991b; Dahl, 1972), is a core causative factor for health authority driven reduction the sodium content of processed foods. The primary source of sodium in processed meats is from salt (NaCl). NaCl is used in the production of meat products because of its effects on texture, flavour and shelf life. Salt reduction in meat products thus has adverse effects on water and fat binding, impairing overall texture and increasing cooking loss, and also on sensory quality, especially taste.

The primary use for salt in meat is its ability to extract myofibrillar protein. Myofibrillar proteins can participate in three classes of interactions: protein–water, protein–lipid and protein–protein (Acton & Dick, 1984), which are characterised by functional properties such as water binding, fat binding and gelation. Their increasing solubility and interactions affect oil binding and water holding ability, stability, viscosity, density and other characteristics of emulsions. An important functional characteristic of proteins is gel forming ability. Myofibrillar proteins extracted using salt during the mixing process denature and associate into a gel when the dispersion is heated. The texture of the processed meat product will depend on the structure of the matrix formed when the proteins gel, the amounts and types of particle and solutes entrapped in the gel matrix and the moisture content of the finished product (Foegeding & Lanier, 1987).

Fats and oils play vital functional and sensory roles in various food products. Fats interact with other ingredients to develop texture, mouth feel and assist in the overall sensation of lubricity of foods (Giese, 1996). The proposed relationships between high cholesterol level and low polyunsaturated/saturated fatty acids (PUFA/SFA) ratio and the rise in coronary heart diseases has resulted in focusing on high fat food products including some meat products (Giese, 1992). Therefore, researchers have been working on strategies to reduce animal fat usage in meat products (Delgado-Pando, Cofrades, Ruiz-Capillas, Solas, Triki & Jiménez-Colmenero, 2011; Özvural & Vural, 2008).

The concept of using food to provide health benefits beyond basic nutrional value to aid in alleviation of illness is becoming more popular with the consumer (Kapsak, Rahavi, Childs & White, 2011). Functional foods are a category of food which has been developed to incorporate functional ingredients and have potential health beneficial effects upon consumption. Many different types of functional ingredients exist and are commercially available, each with their own profile in terms of health promoting capability. One example of such an ingredient is co-enzyme Q10. The therapeutic value of CoQ10 is of interest to many researchers and is often used as a supplement in due to its potential beneficial roles in the prevention and treatment of heart disease, diabetes, Parkinson's disease and its anti-carcinogenic properties

(Lieerman, Lyons, Levine & Myerburg, 2005; Chew & Watts, 2004; Beal, 2002; Langsjoen & Langsjoen, 1999; Lockwood, Moesgaard & Folkers, 1994)

The objectives of this Thesis were to gain an understanding of the European consumers' views on processed meats, in terms of health status and use as a functional food. Following this information, the impact of salt and fat reduction on the overall sensory and physiochemical quality of different comminuted processed meat products were examined using a clean label approach and compared agaibst commercial products. The bioassessability of co-enzyme Q10 in processed meats was also assessed using *in-vitro* digestion studies and HPLC qualitative and Quantitative analysis. Finally the sensory and physiochemical changes caused by the addition of CoQ10 to a processed meat system were determined.

Chapter 2. Literature Review

2.1 Overview of the Meat Industry and Potential Growth

The meat industry is one of the most important global industries, livestock alone comprises a significant global asset with a value of at least \$1.4 trillion (Thornton, 2010). The livestock sector itself is responsible for the employment of at least 1.3 billion people globally and also helps to support the livelihoods of 600 million small farmers in the developing world (Thornton, Jones, Owiyo, Kruska, Herrero, Kristjanson, Notenbaert, Bekele & Omolo, 2006). Meat products have been estimated to contribute 17 per cent to total caloric intake and 33 per cent to protein consumption globally (Rosegrant, Sinha, Alder, Ahammad, de Fraiture & Yana-Shapiro, 2009).

Statistics released for 2010 show the top three producers of all meat are China (80 million tonnes), the United States (42 million tonnes) and Brazil (23 million tones) (FAOSTAT, 2013). China is the leading producer of pork meat (51 million tonnes), while the US the second ranked country in terms of pork production produces 9 million tonnes and Germany which is ranked third produces 4 (FAOSTAT, 2013). The US is the top ranked country in terms of both, chicken and beef, producing 16.3 and 11 million tonnes of each commodity, respectively. China is ranked second for chicken (11.7 million tonnes) and third (6 million tonnes) for beef, whereas Brazil is ranked second for beef (9 million tonnes) and third for chicken (10.7 million tonnes) (FAOSTAT, 2013).

The production of meat was calculated to be 297 million tonnes in 2011, which is an increase of 0.8 percent from 2010 production levels (FAO, 2012). The FAO (2012) predicted that by the end of 2012, meat production would reach 302 million tons.

However, while meat production continues to increase, meat consumption has been negatively impacted upon in recent years by health concerns owing to the composition of fresh and processed meats, animal welfare issues and concerns over the impact of meat production on the environment which have created negative consumer attitudes towards meat (Brewer & Rojas, 2008; Coffey, Mintert, Fox, Schroeder & Valentin, 2005; Garnier, Klont & Plastow, 2003; Thompson, Muriel, Russell, Osborne, Bromley, Rowland, Creigh-Tyte & Brown, 2002).

2.2 Consumer Perception of Meat and Processed Meat

The perception of the meat industry by the consumer is of vital importance as it directly impacts on profitability. While consumers can not be placed into one bracket which describes their influences on purchase, what is known are the most important qualities influencing a consumers' choice in meat and meat products, which include; colour, visible lean, drip loss, texture, juiciness, good overall flavour, freshness, health and safety as well as ethical concerns in terms of sustainability and animal welfare (Acebrón & Dopico, 2000).

Malnutrition remains a major problem in many developing countries, proteinenergy malnutrition in particular is a primary cause of death to children in developing countries with poverty being cited as a primary cause (Müller & Krawinkel, 2005). This intern has made meat in these countries to be seen as a valuable and vital commodity. Alternatively in the developed world consumers are becoming more aware of the correlation between what they eat and their overall well being (Fonseca & Salay, 2008; Angulo & Gil, 2007; Mollet & Rowland, 2002), this in turn impacts what foods they consider healthy to eat. Due to the negative public attention that meat production and consumption has received over the past few decades meat consumption has fluctuated greatly. Consumers have been shown to be distrustful towards meat products and the industry in general (Verbeke, Pérez-Cueto, de Barcellos, Krystallis & Grunert, 2010; Tersteeg, Koolmees & van Knapen, 2002). However,

Processed meats in particular receive a great deal of negative attention form media sources (INH, 2012; HSPH, 2010) which further reinforces negative perceptions. Consumers are often told to avoid all processed foods in favour of a diet that is high in fresh lean meats, vegetables, fruit and whole grains. Many organisations which promote such diets package all processed foods as being the same as fast foods from chain restaurants stating that these foods are high in saturated fat, salt, sugar and toxic chemicals. Processed meats in particular are often seen by the consumer to be an unhealthy product, Tobin, O'Sullivan, Hamill & Kerry, (2013)b found that 50% of European consumers thought of processed meats as an unhealthy product, citing high levels of salt and fat as the root cause.

Consumers must be made aware of the importance and requirement for processed meats manufacture and the ethical necessity to minimise waste generated following animal slaughter. Processed meats allow for utilisation of all the muscles and associated by-products which can be unpalatable in their native forms. It is important to show the consumer that processed meats can be produced in a manner which will satisfy all the important qualities which consumers look for in meat products as well as addressing health concerns in a practical and meaningful way.

2.3 Health and Safety Concerns For Meat and Meat Products

2.3.1 Nutritional Concerns

The role of meat in the human diet varies greatly throughout the globe. In the developing world meat is seen as a source of nutritional calories and an excellent source of protein. In the developed countries of the world meat has become stigmatised as an unhealthy food due to research correlating increased consumption of meat to various non-communicable diseases. The majority of studies focus on the association between meat consumption and obesity, cardiovascular disease (CVD) and cancer.

Cardiovascular disease is one of the largest causes of mortality in the world (Murray & Lopez, 1996). An increased risk of developing CVD has been associated with red meat consumption since the early 1980's (Li, Siriamornpun, Wahlqvist, Mann & Sinclair, 2005). CVD has been estimated to cost the EU economy $\in 169$ billion every year (Petersen, Peto, Rayner, Leal, Fernandez, and Gray, 2005), similar figures have also been shown for the USA with an estimated cost of \$403.1 Billion per year (Thom, Hasse, Rosamond, Howard, Rumsfeld and Manolio, 2006). CVD is often linked with a number of risk factors, which include but are not restricted to hypertension (Morgan, Aubert & Brunner, 2001; Sacks, Svetkey & Vollmer, 2001; Tuomilehto, Lindstorm, & Eriksson, 2001) and obesity (Aggett, Antoine, Asp, Bellisle, Contor and Cummings, 2005).

The primary reason for these associations stems from the high levels of saturated fatty acids naturally present in animal fats, which in the past have been associated with an increased risk of CVD due to their potential effects in raising serum low-density lipoproteins (LDL)-cholesterol (Ascherio, Katan, Zock, Stampfer, Willett, 1997). However, a recent study by Siri-Tarino, Sun, Hu & Krauss (2010) conducted a meta-analysis of epidemiologic studies which estimated the risk of CVD associated with an increased intake of dietary saturated fat and found that there is insufficient evidence to conclude that dietary saturated fat is associated with an increased risk of CVD.

The consumption of processed meat in particular has been associated with and increased incidence of CVD, which highlights the need to separate the health impact of different types of processed versus unprocessed meats in the diet (Micha, Wallace & Mozaffarian, 2010). Higher levels of sodium and nitrate in processed meats were observed by Micha et al. (2010) to be likely contributors to the increased incidence of CVD. Sodium is known to significantly increase blood pressure (He & MacGregor, 2002) and also promote vascular stiffening with regular consumption (Sanders, 2009). Hypertension a term which describes high blood pressure has high a global prevalence. In a review, by pooling data it was estimated that 26 percent of the world adult population (972 million) had hypertension in the year 2000 (Kearney, Whelton and Reynolds, 2005).

Roughly 75% of total dietary intake of sodium in many people's diets has been shown to come from processed foods such as breads, pasta, and processed meats (Appel & Anderson, 2010). The World Health Organisation (1990) recommended a total dietary intake of salt of 5 to 6g/day, however, this figure is greatly exceeded in many industrialised European countries and in the United States. In fact it has been estimated to be as high as 9 to 12 g NaCl/day (Doyle & Glass, 2010; Intersalt Cooperative Research Group, 1988).

Similarly to sodium, a by-product of nitrate called peroxynitrite is also known to contribute to vascular oxidative stress (Förstermann, 2010). A separate study by Wagemakers, Prynne, Stephen & Wadsworth, (2009) found no significant association

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between red or processed meat consumption in 1989 and 1999 and LDL-cholesterol concentrations and blood pressure when measured in 1999. However, the combined intake of red and processed meat in 1999 was found to have a significant positive association with blood pressure but only in males. A recent meat analysis study on the association of red and processed meat consumption and risk of stroke found no association to hemorrhagic stroke; however data did indicate an increased risk of ischemic stroke (Chen, Lv, Pang & Liu, (2012).

Elevated phosphate levels in blood serum have been labelled a risk to people with impaired renal function (Eddington, Hoefield, Sinha, Chrysochou, Lane, Foley, Hegarty, New, O'Donoghue, Middleton & Kalra, 2010) and recently to people with no health issues (Sullivan, Sayre, Leon, Machekano, Love, Porter, Marbury & Sehgal, 2009). Specifically, high phosphate concentration in blood serum is linked with calcification of the arteries in healthy men (Foley, Allan, Collins, Herzog, Ishani, & Kalra, 2009). Most of the phosphate in the human body is bound to calcium and helps form bones and teeth. The remainder is found throughout the body as it aids the kidneys to filter waste and a vital component of ATP, which is the primary compound used for chemical energy transfer in the body.

Restricting the content of natural phosphate in our diets such as organic esters found in food would be pointless as it is not fully absorbed and if intake is too restricted may lead to protein malnutrition (Waterlow & Golden, 1994). However, inorganic phosphate added to foods such as meat products is absorbed well by the body and can lead to elevated phosphate levels in the blood serum (Ritz, Hahn, Ketteler, Kuhlmann & Mann, 2012). Considering phosphates are an important part of the human diet, care must be taken to reduce phosphate where possible, but not eliminate them.

2.3.2 Correlations between Meat Consumption and the Obesity Epidemic

The prevalence of excess levels of adipose tissue build up in the human body is widely recognised as a leading world health problem and is referred to as an obesity epidemic (WHO, 2000). The World Health Organisation (WHO) found in 2008, that more than 1.4 billion adults, aged 20 and older, were overweight. Out of these overweight adults, 200 million men and nearly 300 million women were classified as obese (WHO, 2012). The WHO also found that more than 40 million children under the age of five were overweight in 2010 (WHO, 2012).

Though the mechanisms behind excessive body weight gain are not fully understood, it is often seen that obesity is primarily caused by an energy imbalance between calories consumed and calories expended. Processed meats and poultry have been associated with increased waist circumference in women, however no significant associations were found for men (Halkjær, Tjnneland, Overvad, & Sørensen, 2009). Interestingly, Halkjær et al. (2009) also found that diets high in red meat showed significant reduction in waist size over a five year period for both men and women. Contrary results were published by Wagemakers, et al. (2009) who found red and processed meat intakes in 1989 independently and collectively had a significant positive association with waist circumference increased by 1999. However they concluded that more studies of a wider population are required with more care being taken to accurately estimate actual meat consumption.

Past studies have shown that meat, both poultry and red meat, can be included into a weight loss aimed diet and have equivalent rates of success (Campbell and Tang, 2010; Leslie, Lean, Baillie & Hankey, 2002). In fact, diets including high levels of protein have been shown to help reduce weight (Soenen, Bonomi, Lemmens, Scholte, Thijssen, van Berkum & Westerterp-Plantenga, 2012). Out of the

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macronutrients, protein is considered to be the most satiating and also has the highest diet-induced thermogenesis (Paddon-Jones, Westman, Mattes, Wolfe, Astrup, & Westerterp-Plantenga, 2008).

2.3.3 Meat Consumption and Association with Cancer Risk

Health concerns have been raised about the consumption of meat increasing the risk of developing cancer (Cross, Leitzmann, Gail, Hollenbeck, Schatzkin & Sinha, 2007). Most research in this area tends to focus on colorectal cancer; however the findings of these studies have been inconsistent. It is extremely difficult to identify a specific food or food group's particular impact on cancer development in the human diet due to the varied diets of human subjects in studies (Magalhaes, Peleteiro, & Lunet, 2012).

Researchers have put forward several hypotheses on the reasons why meat consumption increases the risk of colorectal cancers. Bruce, (1987) suggests a possible cause to be the increase in bile production due to the high levels of dietary fat, the surfactant qualities of the bile damage the mucosal membrane which in turn promotes cell death and proliferation. Visek & Clington (1991) suggested that the fermentation of protein in the large intestine produces toxic products which can damage the mucosa. Other suggested causes include genocidal free radicals formed from the presence of iron (Nelson, 2001); the production nitrosamines from added nitrates (Bingham, Pignatelli, Pollock, Ellul, Malaveille, Gross, Runswick, Cummings & O'Neil, 1996); the creation of heterocyclic amines and polycyclic aromatic hydrocarbons during cooking (Sugimura, Wakabayashi, Nakagama, & Nagao, 2004).

However, in relation to fat, protein fermentation and iron these explanations seem unlikely and have shown no significant effect on the risk of developing

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colorectal cancer (Alexander, Cushing, Lowe, Sceurman & Roberts, 2009; Windey, De Preter & Verbeke, 2012; Lee, Anderson, Harnack, Folsom & Jacobs Jr, 2004). Heterocyclic amines and polycyclic aromatic hydrocarbons are likely contributors to colorectal cancer risk; however it is improbable that in the range of normal human exposure that they could be the solitary causative factor (Cross, & Sinha, 2004). Additionally, the corresponding consumption of high fiber and flavonoid-containing foods are recommended as a simple means to reduce the potential carcinogenic exposure from these compounds (Felton & Malfatti, 2006).

Nitrates are added to cured meat in the form of sodium nitrite or cultured celery extract where the naturally present nitrates are reduced to nitrites by a starter bacterial culture (Sebranek, Jackson-Davis, Myers & Lavieri, 2012). Nitrate (NO⁻³) is reduced to nitrite (NO⁻²) and then further reduced to nitric oxide (NO) in food products. The nitrates react with the myoglobin present in meat to form nitrosylmyoglobin which forms the bases of the cured meat colour. Nitrates have been shown to produce nitrosamines which are formed when nitrite is in the presence of low molecular weight amines (Parthasarthy & Bryan, 2012). Nitric oxide (NO) is found throughout the body and its role in the body has been known for many years (Olesen, 2008 and Koss, 1999). Various health disorders have been associated to a deficiency in NO (Moncada, Palmer, & Higgs, 1991). More recently researchers have begun to look at the benefits of nitrates as a therapeutic agent to treat NO insufficiency (Bryan & Loscalzo, 2011; Lundberg, Weitzberg & Gladwin, 2008; Wilkes, 1994). Overall the evidence linking meat to a role in colorectal carcinogenesis is relatively weak and unclear.

2.4 Processed Meat Production and Ingredients

Meats are industrially processed with the addition of various ingredients in order to protect or modify the flavour, colour, texture, juiciness and/or to aid in preservation. Common meat additives include the following; Water; Salt; Nitrates; Phosphates; Stabilizers; Emulsifiers; Flavourings; Seasonings; Sweeteners; Acidifiers; Tenderisers; and Antioxidants. Each ingredient is used to carry out a specific function in the product.

Salt, arguably the most important ingredient in processed meat, is a vital component in many products for its role in preservation, flavour enhancement and especially its influence on water holding capacity (Lawrence, Dikeman, Hunt, Kastner & Johnson, 2003; Silva, Morais, & Silvestre, 2003). Salt's preservative effect in food is brought about by the increase in osmotic pressure in high concentrations and the capacity to decrease the water activity of foods which reduces or completely halts vital microbial processes (Durack, Alonso-Gomez & Wilkinson, 2008).

The distinctive taste of salt is created by the Na⁺ cation in combination with Cl⁻ anion effect on taste receptor cells in the mouth (Miller & Bartoshuk, 1991; Murphy, Cardello & Brand, 1981). Salts formed with either heavier cations or anions such as potassium chloride (KCl) are found to produce more bitter taste in foods (Murphy et al. 1981). Salt is also enhances flavours and has been shown to increase the perceptions of flavours in meat products (Ruusunen, Simolin & Puolanne, 2001).

The ability of a food to trap water within a three dimensional structure is known as the water holding capacity (WHC) (Chantrapornchai & McClements, 2002). Salt is used in meat products to extracted myofibrillar proteins which associate into a gel when heated and subsequently increase the water holding capacity of the meat product (Foegeding & Lanier, 1987). The Cl⁻ ion in salt is more strongly bound to proteins than the Na^+ ion, resulting in an increased negative charge within the proteins. The negative charges then repulse one another which cause swelling of the myofibrillar proteins (Hamm, 1972).

Nitrite is the active ingredient in the curing of meats and is added predominantly as either the sodium or potassium salt. Nitrites primary functions are as an antimicrobial agent as they are considered an irreplaceable ingredient for the prevention of botulinum poisoning from consumption of cured meat by hindering spore germination (Christiansen, Johnston, Kautter, Howard & Aunan, 1973). It is also used as a source of nitric oxide which is necessary to form the distinctive pink colour. Nitric oxide (NO) is formed by the reduction of nitrate (NO⁻³) to nitrite (NO⁻²) which further reduces to NO, the NO then reacts with the myoglobin in meat to form nitrosomyoglobin.

Due to the relatively high toxicity of NO⁻³ (the lethal dose is roughly 22 mg/kg of body weight in humans); a maximum allowed nitrite concentration in meat products is between 100 and 150 ppm (Food Standards Agency, 2008). As mentioned earlier in this review there is a potential for the formation of known carcinogens such as nitrosamines when nitrates are present in high concentrations and the product is cooked at high temperatures (Parthasarthy & Bryan, 2012). The use of ascorbate in cured meat is known to reduce the formation of nitrosamines (Archer, Tannenbaum, Fan & Weisman, 1975), and its use in cured meats is now required by law in the US (Scanlan, 2000).

The main effects from phosphates on meat systems are on the pH, binding of divalent cations and reduction in viscosity. The changes brought about by phosphates are used as a means of improving the WHC, increasing yields and stabilizing meat emulsions, which in turn decrease cooking losses, extends shelf-life and also improves textural and sensory properties (Knipe, 2003; Varnam & Sutherland, 1995).

Phosphates influence the WHC of meat by affecting the pH, which has a direct effect on the WHC (Hamm, 1972). The WHC of meat is lowest at a pH of about 5.0 (Puolanne & Halonen, 2010), deviating from this point will increase the WHC by creating a charge imbalance and increase swelling between proteins (Knipe, 2003). Divalent cations are found in hard water areas as well as naturally present in meat, these cations can reduce the WHC of meat products, this effect however is counteracted by the addition of phosphates which bind the cations. Phosphates effects on viscosity are also an important factor especially for processing. Phosphates are used to decrease the viscosity of the meat emulsion, therefore decreasing the energy needed to mix and chop the meat and fat particles allowing for the creation of a finer emulsion (Knipe, 2003).

Sodium lactate and potassium lactate are added to meat products for their antimicrobial action specifically to inhibit the growth of *Clostridium botulinum* (Maas, Glass & Doyle, 1989). They are also used for their positive affects to the colour and as a flavour enhancer (Miller, 2010). These effects are likely brought about by an increase in pH which delays the oxidation of the red coloured de-oxymyoglobin to a brown coloured metmyoglobin and also reduces the degree of lipid oxidation which can create off flavours in meat.

Flavourings, seasonings and sweeteners are added to meat to offer consumer different flavours and furthers product ranges. Aside form affecting the sensory profile of a product some spices and seasonings such as rosemary; oregano and curcumin to name but a few are known to have antioxidant and antimicrobial effects in meat (Zhang, Xiao, Samaraweera, Lee & Ahn, 2010; Erkan, Ayranci & Ayranci,

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2008; Baratta, Dorman, Deans, Biondi & Ruberto, 1998; Ruby, Kuttan, Dinesh Babu, Rajasekharan & Kuttan, 1995). Antioxidants are used in meat to prevent available oxygen present in surrounding air from producing undesirable changes in the products flavour or colour. Other examples of antioxidants include butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), and Tocopherols (vitamin E).

Stabilisers, emulsifiers or thickeners are added to meat to help maintain a uniform texture. Ingredients such as whey protein concentrate, sodium caseinate, lecitin and a variety of hydrocolloids like gelatine and carrageenan are commonly used in the meat industry for these reasons (Andrès, Zaritzky & Califano, 2006). Cold set binders are of particular interest to the meat industry as then can affect the textural properties of a meat system without the application of heat, therefore keeping the meat product in its natural raw state. Examples of cold set binders include non thermal gelatine, alginate and fibrin (Boles, 2011). Another particular point of interest in cold set binders to the meat industry is its use as a replacement for salt and fat with a goal of creating a more consumer perceived healthy product.

2.5 Reduction of Salt and Fat in Meat Products

2.5.1 Salt Reduction in Processed Meat

Non-communicable disease (NCD), such as heart disease, stroke, cancer, chronic respiratory diseases and diabetes, are slow progressing diseases and seen as a leading cause of mortality in the world. The WHO (2011) estimates that up to 63% of deaths globally are directly attributed to NCD. There is also a substantial economic burden from NCD due to health care cost and loss to productivity, in fact the WHO estimates that the loss in income by 2015 could be as high as 54 billion dollars in India, roughly 6.6 billion for the UK, Russia and Pakistan, and between 10 and 13

billion for Brazil and China (WHO, 2006). Diet is not the only factor which affects our likelihood of developing a NCD, however it is one of the most important factors, because of this health professions, government agencies and dietician recommend that people aim to have a balanced diet containing all food groups.

Consumers are beginning to be influenced by the contribution diet plays in our overall health (Fonseca & Salay, 2008; Angulo & Gil, 2007; Mollet & Rowland, 2002). As consumer attitudes change so must their product choice, therefore the need to develop more healthy versions of products is required. The main source for sodium in our diets comes from salt. The reduction of salt in meat has been an area of great interest due to compulsory decrease in sodium levels of foods by regulatory agencies (FDA, 2010; FSAI, 2010; FSA, 2009).

Numerous methods of reducing salt in meat have been developed over the last few years. Many researchers have looked into the use of alternative to salt using compound with similar technological effects. The use of potassium chloride (KCl) has been suggested as a potential salt replacer (FSAI, 2005), however when used in high concentrations the KCl produces a strong bitter taste (Murphy et al. 1981). The use of KCl as a partial substitute for salt is viewed as a good way to reduce sodium levels (Ruusunen & Puolanne, 2005). Varying mixtures of KCl and NaCl already exist on the market such as Lo[®] salt, Morton Lite Salt[®] and Pansalt[®]. The use of other salts in conjunction with NaCl has been shown to increase the rate of lipid oxidation in meat products (Ripollés, Campagnol, Armenteros, Aristoy & Toldrá, 2011). Guàrdia, Guerrero, Gelabert, Gou & Arnau, (2008) and Jime'nez Colmenero, Ayo & Carballo, (2005) showed that the use of KCl/NaCl mix with other ingredients such as potassium lactate, casemate and flavouring maskers were able to produce frankfurter and sausage type products with similar sensory profiles as products only made with NaCl. A study by Champagne, Fontaine, Dussault & Delaquis, (1993) found using KCl as a partial replacement for NaCl inhibited the growth of fermentative bacteria in pork products which retarded fermentation however this effect could be avoided by the use of a properly selected starter culture. Other salts such as magnesium chloride can also be used, however NaCl remains the most proficient salt at delivering the most consumer acceptable taste and is the most efficient salt at myofibrillar protein extraction (Munasinghe & Sakai, 2004). López-López, Cofrades, Ruiz-Capillas & Jiménez-Colmenero, (2009) reported that the use of seaweed could also be used as a means to achieve salt reduction in frankfurters. Other ingredients such as milk proteins have also been reported as potential salt replacers in meat products (Hayes, Desmond, Troy, Buckley & Mehra, 2006).

The use of phosphates has been shown to be effective at lowering the salt requirement for processing in meat products (Barbut, Maurer & Lindsay, 1988; Puolanne & Terrell, 1983). Ruusunen, Vainionpää, Lyly, Lähteenmäki, Niemistö, Ahvenainen & Puolanne, (2005) found that meat patty firmness can be maintained with a reduction of 40% NaCl if phosphates are used. Puolanne & Ruusunen, (1980) reported an attainable 0.3-0.5% salt decrease in sausages with the addition of phosphates.

The use of flavour enhancers or masking agents such as yeast extracts; monosodium glutamate or lactates are used in conjunction with salt to compensate for the reduction in salt intensity (Brandsma, 2006). Ruusenen, et al. (2001) and Pasin, O'Mahony, York, Weitzel, Gabriel & Zeidler, (1989) reported the effectiveness of such products in sausage style meats. The use of taste blockers such as adenosine 5'monophosphate can also be used to reduce the bitter taste created by the KCl in salt mixtures (McGregor, 2004).

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Another strategy employed by the food industry is the optimisation of salts physical shape. This strategy centres on increasing the surface area of the salt crystals used in manufacturing (Desmond, 2006). Alternate processing procedures have also been reported to be effective methods to reduce salt. Crehan, Troy & Buckley (2000) found the use of high pressure treatment could be used as an effective method to reduce salt in frankfurters.

2.5.2 Fat Reduction in Processed Meat

Fat plays an important role in processed meat for both flavour and texture. The use of fat replacers can drastically change the sensory profile of a meat product. Over half the volatile compounds found in meat originate from the lipid fraction (Brewer, 2012). Fat has a similarly large impact in terms of eating quality for meat products as they interact with other ingredients and help to develop texture, mouthfeel and provide a lubricating effect (Webb, 2006; Javidipour, Vural, Ozbas and Tekin, 2005; Wood, 1990).

Fat reduction in meat is achievable using similar techniques as with salt reduction. Most of the fat reduction studies focus on the reduction of saturated fats rather than a reduction in total fat as these fats are seen as unhealthy by many consumer groups. The use of unsaturated oils such as vegetable or fish oils as a partial substitute to animal fat in meat products has received a great deal of attention. Özvural & Vural, (2008) reported that the use palm oil, palm stearin, cottonseed oil, hazelnut oil and mixes of these oils could be used to increase the ratio of unsaturated to saturated fatty acids and maintain product quality. Similar findings are reported by Delgado-Pando, Cofrades, Ruiz-Capillas, Solas, Triki & Jiménez-Colmenero, (2011) for olive oil, linseed oil and fish oil mixes. Both these studies report a change in certain physiochemical properties of the meat products, however the products scored as acceptable in consumer trials.

Other non lipid ingredients such as protein based substitutes have been reported to be suitable fat replaces for meat products. For example, Sampaio, Castellucci, Pinto e Silva & Torres, (2004), Cengiz & Gokoglu, (2007) and Murphy, Gilroy, Kerry, Buckley & Kerry, (2004) reported findings on the effectiveness of fat reduction using whey protein, soy protein and surimi respectively. As with oil blend, textural differences were detected in the finished product but these changes did not have a negative impact on overall sensory acceptability.

The use of carbohydrate and gum based fat replacers have also been studied. Ayo, Carballo, Solas & Jiménez-Colmenero, (2008), reported consumer acceptable low fat products produced with walnut. Also, García-García, & Totosaus (2008) found starch added with either K-carrageenan or locust bean gum could produce similar results in terms of texture as full fat products.

2.6 Health Benefits from meat consumption

Often under-reported by the media are the many health benefits associated with meat consumption. Meat contains a high concentration of biologically valuable protein. Animal proteins contain all the essential amino acids required by the human body, unlike whole grains and cereals which tend to be limited in lysine and threonine (Block, 1945). Protein plays a vital role in the body and many researchers are looking into high protein diets as a means to prevent and manage non communicable disease. High protein diets have been linked to offsetting age related muscle loss (Phillips, 2012); prevention of stunting in developing countries (Krebs et al. 2011); and weight reduction (Weigle, Breen, Matthys, Callahan, Meeuws, Burden & Purnell, 2005).
However, when taken to the extreme or not properly balanced, individuals who use high protein diets can lead to inadequate vitamin and mineral intake, which can develop into potential cardiac, renal, bone, and liver problems. (St. Jeor, Howard, Prewitt, Bovee, Bazzarre & Eckel, 2001).

The fatty acid composition of meat varies by animal, age, sex, breed, diet and even the cut of meat used (Wood & Enser, 1997). Meat consists of both saturated and unsaturated fatty acids, the ratio of which has become a point of interest to the scientific community. The ratio of unsaturated fat to saturated fat (UF:SF) in beef is roughly 0.11; in lamb 0.15; and in pork 0.58 (Enser, Hallett, Hewitt, Fursey & Wood, 1996). Past meta analysis studies have shown a positive health correlation with an increase in the dietary ratio of UF:SF in regards to reduction in cholesterol (Scollan, Choi, Kurt, Fisher, Enser & Wood, (2001).

The predominant polyunsaturated fatty acids found in meat are the essential n-6 linoleic acid and n-3 alpha-linolenic acid (McAfee, McSorley, Cuskelly, Moss, Wallace, Bonham & Fearon, 2010). In particular n-3 polyunsaturated fatty acids are associated with positive effects on heart health; improving the aggregation of platelets, reduces risk of developing thrombosis, positive effects to the central nervous system, retinal function and the immune system (Siddiqui, Harvey & Zaloga, 2008; Mann, Pirotta, O'Connell, Li, Kelly & Sinclair, 2006; Ruxton, Reed, Simpson, & Millington, 2004).

Conjugated linoleic acid (CLA) are group of isomers of linoleic acid which are found most commonly in the meat and dairy products derived from ruminant animals (Schmid, Collomb, Sieber & Bee, 2006; Turpeinen, Mutanen, Aro, Salminen, Basu & Palmquist, 2002). CLA has garnered attention due to its potential anti carcinogenic

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properties as well as beneficial effects towards hypertension, atherosclerosis and diabetes (Bhattacharya, Banu, Rahman, Causey & Fernandes, 2006).

Meat is also an extremely valuable source for vitamin B12, niacin, vitamin B6, iron, zinc and phosphorus, riboflavin, pantothenic acid, selenium and potentially vitamin D as well as a multitude of endogenous antioxidants and bioactive substances such as; taurine, carnitine, carnosine, ubiquinone, glutathione and creatine. (McAfee et al. 2010; Williams, 2007; Jimenez-Colmenero, Carballo, & Cofrades, 2001).

The high levels of iron in meet is of particular interest as studies conducted around the world have shown significant portions of the population to be deficient in iron (Fricker, Lemoel & Apfelbaum, 1990). The predominant cause of iron deficiency has been linked to menstruational bleeding in women (Milman & Kirchoff, 1992; Fricker et al. 1990). This inadequacy in iron can was shown to be easily prevented with the consumption of readily available heme iron present in red meat (Leonhardt, Kreuzer & Wenk, 1997). Meat is also known to enhance the bioavailability of nonheme iron in foods, making meat both a source and an enhancer for the intake of iron (Mulvihill & Morrissey, 1997).

Bioactive peptides are peptides derived in food that exert a physiological hormone-like effect in humans beyond that of their nutritional value. They are found in a variety of foods such as milk, egg, meat, fish and numerous types of plants. When ingested, bioactive peptides generally confer beneficial effects on the body by either entering the circulatory system through the intestine or by producing local effects in the gastrointestinal tract. These peptides can produce numerous effects in the body depending on their individual amino acid make up, examples include; opiate-like, mineral binding, immunomodulatory, antimicrobial, antioxidant, antithrombotic, hypocholesterolemic, and antihypertensive actions (Hartmann & Meisel, 2007; Kitts & Weiler, 2003; Kovacs-Nolan, Phillips & Mine, 2005; Meisel, 1997). A large amount of peptides and free amino acids are produced during meat processing through proteolysis mechanisms (Toldrá, 2006). Peptides can also be obtained industrially from meat protein wastes (trimmings, organs, collagen, hemoglobin) with the use of commercial proteases (Vercruysse, Van Camp, & Smagghe, 2005). Hundreds of peptides are available from meat, however only a few are bioactive and provide health benefits to the consumer. These bioactive peptides have been linked to the inhibition of the angiotensin I-converting enzyme (ACE) which has been shown to be an effective way to reduce blood pressure (Ahmed & Mugurama, 2010). Peptides with antioxidant effects have also been isolated aswell as antimicrobial peptides (Di Bernardini, Harnedy, Bolton, Kerry, O'Neill, Mullen & Hayes, 2011).

2.7 Meat as a Functional Food

The concept of medication through food is becoming increasingly important to consumers in developed countries (Kapsak, Rahavi, Childs & White, 2011). The desire to create a medicating food lead to the development of functional foods back in the 1980's in Japan, where the name functional food was first coined. Functional food is simply the most common name however many other names such as nutraceuticals, designer foods, and pharmafoods exist which all describe foods that can prevent or treat diseases (Goldberg, 1994). For a food to be considered functional it must fit 3 basic criteria: 1). To be a food or food ingredient derived from natural occurring ingredients and not be ingested as a powder, tablet or capsule; 2). To be consumed daily as part of an overall diet; 3). To have a positive impact on an individual's health, physical performance or state of mind, in addition to its nutritive value (Goldberg, 1994).

The market for such foods is rapidly building and is estimated to continue well into the future. (PWC, 2009; Fern, 2007; Verbeke, 2005). The market has been estimated to reach up to \$130 billion for functional food and drinks world-wide by the year 2015 (Global Industry Analysts, 2010). The acceptance of functional foods has been shown to vary greatly depending social, economical, geographical, political, cultural and ethnicity of the consumer (Verbeke, 2005; Jimenez-Colmenero et al. 2001). To date the majority of the market has been dominated by the dairy industry (Leatherhead Food Research, 2011), however there is still room for the meat industry to emerge as a leading contributor of functional foods.

The improvement of a foods nutritional quality is not a straight forward task as this improvement must be conducted without drastically affecting the consumers need for quality, convenience and price. The majority of functional foods are created through the fortification of a food ingredient using what are called biologically active compounds or bioactive. To choose a suitable bioactive certain factors must be considered such as; is the bioactive under consumed by the target population; how efficiently does the bioactive work in humans: the bioactive's physiochemical properties; will the addition of the bioactive deteriorate food quality; and is it economically available and safe to use.

Different fortification strategies must also be considered. For meat or dairy products strategies include the use of feed supplementation with bioactive compounds in the target animal's diet or the direct addition of bioactives to a processed meat or dairy product. Many recent studies have been conducted on both these strategies (Khan, Arshad, Anjum, Sameen, Waqas & Gill, 2011; Zhang, et al. 2010). Other ingredients There are a multitude of bioactive compounds available with more being produced every year. Goldberg (1994) identified 12 broad groups of substances which

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could be considered bioactive these include: 1). Dietary Fibre; 2). Oligosaccharides;
3). Sugars; 4). Amino acids; 5). Glucosides; 6). Alcohols; 7). Isoprenes and vitamins;
8). Choline; 9). Lactic acid bacteria; 10). Minerals; 11). Unsaturated fatty acids; 12).
Others not including previously mentioned. An example of one is coenzyme-Q10 (CoQ10).

2.8 Coenzyme Q10

Coenzyme Q is a ubiquitously occurring quinone, thus garnering its name ubiquinone. Quinones are a homologous group of compounds found in many living organisms such as animals, plants and yeasts (Turunen, Olsson & Dallner, 2004 and Battino, Ferri, Gorini, Federico Villa, Rodriquez Huertas, Fiorella, Genova, Lenaz & Marchetti, 1990). The most abundant form in humans and animals is Coenzyme Q10 (CoQ10), which contains 10 isoprenoid units in the side chain (Lester and Crane, 1959). CoQ10 is a redox molecule and is found in the body in both a reduced state known as ubiquinol-10 and an oxidised state called ubiquinone-10 (Kubo, Fujii, Kawabe, Matsumoto, Kishida, & Hosoe, 2008).

CoQ10 was first isolated from beef heart mitochondria back in the 1957 in an experiment to determine the mitrochondrial electron-transfer system (Parvst, Žmitek & Žmitek, 2010). This led to the eventual discover of its role in mitrochondrial electron transfer within cells (Mitchell, 1976). CoQ10 plays a key role in the transfer of energy from carbohydrates and lipids into the production of Adenosine triphosphate (ATP), which is often referred to as the "molecular unit of currency" of intracellular energy transfer and is an essential component of respiration (Knowles, 1980). In its reduced form ubiquinol-10 is known to be an extremely effective antioxidant (Kubo et al. 2008). Ubiquinone is reduced to ubiquinol by enzymatic

action prior to absorption in the small intestine (Mohr, Bowry & Stocker, 1992). In this form ubiquinol loosely hold two electrons which it can give up to neutralise free radicals (Kumar, Kaur, Devi & Mohan 2009).

There is substantial interest in the therapeutic value of CoQ10 and it is often used as a supplement in conjunction with standard medical therapy especially when used to treat cardiovascular and neurodegenerative diseases (Overvad, Diamant, Holm, Holmer, Mortensen & Stender, 1999; Langsjoen & Langsjoen, 1999; Beal, 2002). CoQ10 has also been associated with a variety of other health benefits such as potentially aiding sufferers of diabetes (Chew and Watts, 2004; Palacka, Kucharska, Murin, Dostalova, Okkelova, Cizova, Waczulikova, Moricova & Gvozdjakova, 2010); anti-carcinogenic properties (Lockwood, Moesgaard & Folkers, 1994); delaying the onset of Parkinson's disease (Lieberman, Lyons, Levine & Myerburg, 2005 & Shults, Oakes, Kieburtz, Beal, Haas, Plumb, Juncos, Nutt, Shoulson, Carter, Kompoliti, Perlmutter, Reich, Stern, Watts, Kurlan, Molho, Harrison & Lew, 2002); reduces the risk of pregnant women developing pre-eclampsia (Teran, Hernandez, Nieto, Tavara, Ocampo & Calle, 2009), and as a recovery aid after cardiac surgery (Rosenfeldt, Marasco, Lyon, Wowk, Sheeran, Bailey, Esmore, Davis, Pick, Rabinov, Smith, Nagley & Pepe, 2005; Rosenfeldt, Pepe, Linnane, Nagley, Rowland, Ou, Marasco & Lyon, 2002).

CoQ10 is obtained by humans and animals through both endogenous and exogenous sources. It is biosynthesized in the body and is found most concentrated in organs like the heart, kidneys, liver, muscle, pancreas, and thyroid gland. However, as people age the CoQ10 content in these organs decreases and may lead to a deficient level (Kalen, Appelkvist and Dallner, 1989). A deficiency in CoQ10 has been shown to cause dilated cardiomyopathy (DCM) and hypertrophic cardiomyopathy (HCM) due to defective levels of certainc oxidative phosphorylatory enzyme activities (Marin-Garcia, Goldenthal, Pierpont & Ananthakrishnan, 1995; Rustin, Lebidois & Chretien, 1994).

There are many natural sources of CoQ10 in the human diet, the richest being meat, poultry, fish and rapeseed oil (Stocker, 2007, Mattila, & Kumpulainen, 2001) These level however are still quite low (Weber, Bysted & Holmer, 1996), in fact Kumar et al. (2009) have estimated the total dietary intake of CoQ10 only 2-5 mg/day for the average human adult living in the developed world. Considering the recommended therapeutic level of CoQ10 is given as 100-200 mg/day with an upper tolerance limit in-terms of safety as 2400 mg/day this is far too low to achieve beneficial effect in the body (Hathcock & Shao, 2006).

Chapter 3. European Consumer Attitudes on the Associated Health Benefits of Neutraceutical-Containing Processed Meats using Co-Enzyme Q10 as a Sample Functional Ingredient

This Chapter is in the form of a manuscript submitted for publication in *Meat Science* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). European Consumer Attitudes on the Associated Health Benefits of Neutraceutical-Containing Processed Meats using Co-Enzyme Q10 as a Sample Functional Ingredient

Abstract

The meat industry is an important sector of the agricultural economy of Europe. However, the sector has suffered from both economic instability and negative consumer attitudes towards meat products. This study aims to accumulate European consumers' attitudes towards processed meats and their use as a functional food. A survey was set up using an online web-application to gather information on European consumers' perception. Over 500 responses were obtained and statistical analysis was carried out using a statistical software package. Data was summarized as frequencies for each question and statistical differences were analyzed using the Chi-Square statistical test. A significance level of 5% (P<0.05) was set for all analysis. The majority of consumers' were found to view processed meat as an unhealthy product. Most believe processed meats contain large quantities of harmful chemicals, fat and especially salt. Consumers were found to be very pro-bioactive compounds in yogurt style products but unsure of their feelings on the idea of them in meat based products, which is likely due to the lack of familiarity to these products. Many of the respondents were willing to consume meat based functional food products but were not willing to pay more for them.

3.1 Introduction

The manufacture of processed meat products constitutes an important activity sector within the meat industry as its fundamental objective is to convert less commercial and usable forms of raw muscle into more economical and palatable products. There have been many studies in the area of consumer perception of quality of fresh meats (Brunsø, Bredahl, Grunert & Scholderer, 2005; Grunert, 1997, 2005; Grunert & BechLarsen, 2004), however, very little consumer-related research has been conducted in the area of processed meats.

In recent years meat production and consumption has suffered from a great deal of negative publicity. Several major issues of public health concern associated with meat production, namely; the emergence of bovine spongiform encephalopathy (BSE) in the late 1980's, the outbreak of foot and mouth in the 2000's and the illegal use of anabolic steroids as growth promoters caused both economic set back to the industry and created a great deal of mistrust among the public (Coffey, Mintert, Fox, Schroeder & Valentin, 2005; Marsh, Schroeder & Mintert, 2004). This, coupled with concerns pertaining to; meat production impact on the environment (Aguiar, Vieira, Ferreira & de Barcellos, 2008), food product choice including welfare issues (Krystallis, de Barcellos, Kügler, Verbeke & Grunert, 2009; Vanhonacker, Verbeke, Van Poucke & Tuyttens, 2008; Brom, 2000) and negative health issues associated with the consumption of meat, in particular processed meats, has further impacted negatively on the public perception of meat and its consumption.

However, even with all of these negative associations, meat consumption globally continues to increase and is estimated to double from 2000 to 2050 (Steinfeld, Gerber, Wassenaar, Castel, Rosales & de Haan, 2006). This consumption will be both in the form of fresh and processed product. Irrespective of the desire to consume fresh meat over processed products, it is imperative that the consumer is brought to understand that the ethical slaughter of animals for meat production is only justified morally by utilising all muscle and associated by-products derived from butchering; a large proportion of which in its native form is unpalatable, tough, unusable etc. Additionally, the tradition and heritage associated with certain cultural processed meat products will ensure that specific product types and formats prevail into the future, thereby requiring the use of specific meat cuts.

While meat and meat products have received more than their fair share of negative publicity in terms of health and nutrition (often driven by a badly informed media or by those with a vested interest or ethical viewpoint which is negative towards meat production and consumption), it is important that a balanced position for meat in the diet be presented to consumers. Both positive and negative nutritional attributes are associated with meat and meat-derived products. Meat is an excellent source of high biologically valuable protein, contains vitamin B12, niacin, vitamin B6, iron, zinc and phosphorus. Meat is also a source of long-chain omega-3 polyunsaturated fats, riboflavin, pantothenic acid, selenium and possibly also vitamin D and important associated bioactives. Fresh meat is mostly low in fat and sodium and a source of multiple endogenous antioxidants and other bioactive substances including; taurine, carnitine, carnosine, ubiquinone, glutathione and creatine. (Jimenez-Colmenero, Carballo, & Cofrades, 2001; Williams, 2007).

The unhealthy aspects that consumers associate with meat and meat products include; high levels of saturated fat; cholesterol; high levels of sodium and fat (Whitney & Rolfes, 2002). However, these negative attributes that consumers believe are in certain instances quite skewed. For instance, meat is often seen by the consumer as being high in saturated fats compared to other foods. In actual fact, less than 50%

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of lipids found in meat are comprised of saturated fatty acids and of these, only 25– 35% have atherogenic properties. In fact up to 70% (beef 50–52%, pork 55–57%, lamb 50–52%, chicken 70%) of the lipids in meat are unsaturated fatty acids (Romans, Costello, Carlson, Greaser, & Jones, 1994). The same levels, or higher, of saturated fatty acids can be found in many dairy products. However, these products do not receive the same negative attention as that attributed to meat products. In fact, dairy products have been marketed so successfully that the consumer does not associate any negative attributes with such products as they are perceived by the consumers as healthy carriers of nutrition. The issue of cholesterol in meat is also interesting. Previous research (Barton, Marounek, Kudrna, Bures, & Zahradkova, 2007; Muchenje, Hugo, Dzama, Chimonyo, Strydom, & Raats, 2009) has shown that cholesterol in meat is feed and breed-dependent and some meat can contain almost no cholesterol at all.

The use of chemicals such as; sodium chloride, phosphates, nitrates etc. in processed meats are another concern for consumers. Nitrate and nitrite usage, and levels, in meat have been scrutinized severely since the 1970's after it emerged that they can interact with secondary amines to form N-nitrosamines which have carcinogenic properties. However, Cassens (1997) suggested a need to review the effects that residual nitrite levels in cured meats have on health. While numerous studies (Peters, Preston-Martin, London, Bowman, Buckley & Thomas 1994; Preston-Martin & Lijinsky, 1994; Sarasua & Savitz, 1994; Preston-Martin, Pogoda, Mueller, Holly, Lijinsky & Davis, 1996) have reported on the negative health implications of consuming cured meats, it is interesting to note that as more research has been carried out in the area over the past ten years or so, it has emerged that there are also positive potential health benefits associated with the consumption of meats containing nitrite

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and/or reaction products (Jimenez-Colmenero et al., 2001; Toldrá, 2002; Butler & Feelisch, 2008; Powlson, Addiscott, Benjamin, Cassman, de Kok, Van Grinsven, L'hirondel, Avery & Van Kessel, 2008; Hord, Tang & Bryan, 2009; Lansley, Winyard, Fulford, Vanhatalo, Bailey, Blackwell, DiMenna, Gilchrist, Benjamin, & Jones, 2011; Masschelein, Van Thienen, Wang, Van Schepdael, Thomis & Hespel1 2012). There have also been significant strides made in the area to reduce high salt levels in processed meats as it has been associated with public health issues (Tobin, O'Sullivan, Hamill, & Kerry, 2012ab, 2013; Ruusunen, Vainionpaa, Lyly, Lahteenmaki, Niemisto & Ahvenainen, 2005; Morton Salt. 1994; Riera, Martinez, Salcedo, Juncosa & Sellart, 1996; Pasin, O'Mahony, York, Weitzel, Gabriel & Zeidler, 1989; Maurer, 1983).

Recent studies in meat consumption have shown that consumers are being influenced by health and nutritional considerations more and more (Angulo & Gil, 2007; Fonseca & Salay, 2008). Studies have shown that consumers want food to be more than just a necessity to satiate hunger and provide basic nutritional requirements, but as a product that can provide extra benefits, like that delivered by functional foods. In this context, it is critically important to understand how consumers view meat and meat products in their abilities to deliver such benefits.

A food product may be considered as functional if it is derived from naturallyoccurring ingredients; consumed daily as part of an overall diet and provide health benefits beyond basic nutrition (Jimenez-Colmenero, Carballo & Cofrades, 2001). Improving the nutritional quality of food is a difficult task because this improvement must be made without dramatically affecting the consumers need for quality, convenience and price. The global market potential for functional foods and beverages has been estimated to be worth \$130 billion by 2015 (Global Industry Analysts, 2010). The dairy industry has dominated the functional foods market (Leatherhead Food Research, 2011) to date, by solely and carefully promoting dairy products on the basis of their positive health attributes, and further building on this positivity, by using the products to carry other benefits in the form of functional foods, most notably through the use of functionalised yoghurt-styled products. There is, however, no reason why meat and meat products should not be marketed in the same way or be perceived and function as functional foods. In fact, from a global perspective, it probably makes more sense for muscle-based products to be presented to the population as functional foods than dairy products as their widespread consumption is regionally broader than that of dairy products and as stated previously, muscle-based food consumption is anticipated to double in consumption over the next 40 years to levels higher than those predicted for dairy products.

Meat products can be easily functionalised through the natural and dietary supplementation of animal diets using bioactives, neutraceuticals, vitamins etc. (through incorporation of active substances into muscle) or by direct addition of such materials into meat products during processing. While functional meat products can be presented to consumers in different ways, what is not clear is how consumers might react to them generally and in processed meat formats.

There are many different bioactive compounds available on the market today and the listing increases annually. An example of one bioactive of interest is Q10 (CoQ10). Scientists have been researching CoQ10 for many years because of its importance in the human body and the potential health benefits it can offer (Rosenfeldt, Marasco, Lyon, Wowk, Sheeran, Bailey, Esmore, Davis, Pick, Rabinov, Smith, Nagley & Pepe, 2005; Chew & Watts, 2004; Rosenfeldt, Pepe, Linnane, Nagley, Rowland, Ou, Marasco & Lyon, 2002; Lockwood, Moesgaard & Folkers, 1994). CoQ10 is a perfect example of a functional bioactive that could be incorporated into processed meat products because it offers protection from cardiovascular and neurodegenerative diseases (Overvad, Diamant, Holm, Holmer, Mortensen & Stender, 1999; Langsjoen & Langsjoen, 1999; Beal, 2002).

Therefore, the objectives of this study were to assess consumer attitudes to processed meats generally and determine their views towards using processed meats as functional foods through their carriage of bioactive substances like that of CoQ10.

3.2 Materials and Methods

3.2.1 Questionnaire Preparation

An online survey was carried out in relation to consumers' perception of processed meat products and their attitudes towards bioactive compounds in meat products, with particular attention paid to Co-enzyme Q10. The survey was kept short, in total it consisted of nineteen questions, this was to encourage consumer participation. At the beginning of the questionnaire participants were informed that its purpose was to determine "consumer attitudes towards processed meat and bioactive compound addition", a brief description of bioactives was also given to respondents "Bioactives are compounds which have actions in the body that may promote good health". Examples of bioactives compounds were given as the commonly known "plant sterols" and "probiotics".

3.2.2 Research Questions

The first page of the questionnaire obtained some basic background information which included the age, gender and a rough estimate of the amount of processed meat the respondent consumes. The second page of the questionnaire contained 6 questions focusing on consumer attitude towards processed meats. See Table 1 for questions asked on page 2.

Respondents were asked to rate each question on the basis of whether they; Strongly Agree; Agree; Unsure; Disagree; Strongly Disagree.

Page three of the questionnaire looked at the attitudes of consumers towards bioactive compounds and contained the questions shown in Table 2.

Respondents were required to answer each question as one of the following: Extremely Like; Like; Unsure; Dislike; Extremely Dislike. Page four questioned consumers' knowledge of Co-enzyme Q10. The question which were asked and the responses respondents gave are shown in Table 3.

The final page in the questionnaire contained two finishing questions which looked at consumer attitude towards the consumption and purchase of meat based products with the addition of Co-enzyme Q10, these questions are presented in Table 4.

3.2.3 Evaluation of the Questionnaire

The questionnaire was distributed online through the use of social media websites, university mailing lists and survey mailing lists. To maintain control of only examining European consumers' attitudes any completed questionnaire from a computer IP address registered outside of Europe was not included. Completed questionnaires were coded into a Microsoft[®] Excel worksheet and transferred into IBM SPSS Statistics 20 for Windows (SPSS, Chicago, IL, USA) software package to carry out statistical analysis. The confidence interval of 95% for populations exceeding 1,000,000 individuals was given as minimum 384 respondents however for large populations this is usually rounded up to 400 (Survey Monkey, 2013). Data was summarized as frequencies for each question and statistical differences were analyzed using the Chi-Square statistical test. A significance level of 5% (P<0.05) was set for all analysis.

3.3 Results and Discussion

3.3.1 Demographic

A total of 548 respondents completed the survey. Of these respondents 70% were female and 30 percent male (Table 5). Age was evenly distributed between the ages of 21 and 55 (Table 5), adults aged 18 to 20 only accounting for 2.5% of the total respondents and adults over 56 accounted for roughly 11%.

Level of processed meat consumption amongst respondents found that majority of respondents (68%) consume processed meat products at least once a week (Table 5), with almost 40% of respondents consuming meat multiple times over the course of a week and once a week for another 30%. Fewer than 5% reported consuming a processed meat product daily and another 4.74% claimed to never eat processed meats. The remaining 21% of respondents indicated that they rarely consume processed meats.

3.3.2 Consumer Attitudes towards Processed Meat

Less than 1% of the total respondent (n=548) strongly agreed and only 15.7% agreed that processed meats were perfectly healthy to eat (Table 6). Most people disagreed with 47.63% disagreeing and a further 14% strongly disagreeing, 19.5% of people were unsure. Respondents aged between the ages of 36 to 40 disagreed statistically (P<0.05) more than respondents aged 56 to 60 (Table 6). There was a much more varied opinion with respondents aged 56 and older. Men were found to statistically (P<0.05) agree that processed meat were healthy when compared to women, who were statistically found to more likely strongly disagree (Table 6), a higher negative attitude towards meat in women has also been shown in work by Kubberod, Ueland, Rodbotten, Westad & Risvik (2002). The importance of consumer gender on attitude has also been pointed out in several studies (Dennison & Sheperd, 1995; Sheperd, 1988)

The level of consumption of processed meat products had a great influence on consumer perception, as expected respondents who ate processed meats more regularly were statistically (P<0.05) more likely to agree than respondents who rarely or never consume such products, with almost 90% of respondents who never eat processed meat disagreeing or strongly disagreeing.

When asked if processed meats were unhealthy to eat, respondents generally answered almost inversely to the previous question. Over 50% either strongly agreed or agreed with the statement (Table 6). Less than 2% strongly disagreed and 16.79% disagreed, as with the previous question 20% remained unsure. No age group was found to disagree or agree statistically more than the others, however men were shown to statistically (P<0.05) agree less and disagree more than there female counterparts (Table 6). The frequency of processed meats consumption continued to influence respondents feelings on healthiness with statistical differences (P<0.05) found between those who report eating processed meats often compared to those who rarely do (Table 6).

Respondent's views on the healthiness of processed meats were slightly different when addressing if they were healthy as part of a larger balanced diet. The majority believed them to be so with just under 50% either agreeing or strongly agreeing, roughly 20% still remained unsure, and the final 30% disagreeing (Table 6). Once again, no particular age group was found to disagree or agree statistically more than the others. Men were found to statistically (P<0.05) strongly agree more than women and statistically (P<0.05) disagree more (Table 6). At least 50% of respondents who eat processed meats at least once a week tended to agree or strongly agree, almost 80% of those who report eating processed meats daily agreed or strongly agreed. In comparison to question 4 by replacing "perfectly healthy product" with "healthy as part of a larger balanced diet" regular consumers answers differ dramatically, however people who never eat processed meats are stead fast in there beliefs that processed meats are an unhealthy product.

To grasp consumer perception of processed meat ingredients, respondents were asked if they agree or disagree with statements which said processed meats were full of harmful chemicals, fat and salt. 50% of respondents agreed or strongly agreed that processed meats were full of harmful chemicals with almost 30% unsure, more respondents had a perception of high levels of fat with 66% either strongly agreeing or agreeing, less answered unsure (20%) as compared to how they perceived chemical content (Table 6). Salt content perception however, showed that consumers were much less unsure with only 8% answering so. Almost 80% of consumers believed that processed meat products were full of salt, far greater than for fat and chemicals, only

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2% disagreed with this statement and nobody strongly disagreed. Consumer concerns over their salt intake has been highlighted in work by Grimes, Riddell & Nowson (2009), who found that 44% of participants in their study had concerns over intake of salt and 46% believed that reducing salt intake would improve their health.

No age group contained any significant increase or decrease in numbers agreeing, however women yet again showed more distrust towards processed meats, with significantly (P<0.05) more women strongly agreeing (Table 6) with all three statements compared to men answered as significantly (P<0.05) disagreeing for both salt and harmful chemicals. Male respondents were twice as likely to respond as unsure when answering the statement about salt content compared to women. This further shows women's higher criticism of meats than their male counterparts. Respondents who never consume processed meats were by far the harshest critics, over 90% either agreed or strongly agreed that processed meat was full of chemical and salt and over 70% for fat, this was significantly (P<0.05) higher than those who consume these product once or more a week.

Twigg (1979), postulated that meat resentment arises from certain key feature, which included appearance of blood within the meat, redness and origin, others have concluded that white meat like chicken is more favourable than red meats due to the white colour being less associated with blood or living animals (Gregory, 1997; Guzman & Kjærnes, 1998). This survey however suggests that a further dislike for and criticism for processed meat products has arisen from the consumers perception of the ingredients contained within the products, like caused by the growing health and nutritional concerns of consumers (Resurreccion, 2003).

3.3.3 Consumer Attitudes towards Bioactives

Childs (1997), claimed consumer acceptance was a key factor in determining the success of bioactives in food. Participants in this study generally had a favourable attitude towards bioactives. A majority of almost 60% answered extremely like or like when asked on their thoughts about bioactives, 32% answered unsure and only 8% reported a dislike (Table 7). Previous studies have shown attitudes towards functional foods to be influenced by a number of factors such as; age, gender and education (Bhaskaran & Hardley, 2002; Bower, Saadat & Whitten, 2003; de Jong, Ocke', Branderhorst & Friele, 2003; Urala & La"hteenma"ki, 2004; Verbeke, 2005). This study however found no statistical differences in attitudes between age, gender and level of processed meat consumption.

When asked on their attitudes towards bioactives in particular products, respondents were found to be clear of their liking in the more common yogurt style products with 60% liking or strongly liking (Table 7). Only 7% disliked the idea and the remaining were unsure how they felt. For the less commonly available meat based product with bioactives, the majority of respondents answered unsure (60%), and more than twice as many maintained a dislike for the idea (18%) (Table 7). This finding is in agreement with work done by Urala & Lähteenmäki (2003) who found that consumers do not perceive functional food as a homogeneous food group but a diverse category of foods items. As with the previous question no statistical differences where found between age and gender of participants, however respondents who consumed processed meat more often statistically (P<0.05) strongly disliked the idea of bioactives in meat and yogurt less than those who rarely or never ate processed meats (Table 7).

The final question on page 3 queried respondents' thoughts on whether meat was a suitable carrier for bioactives. The majority answered unsure (> 60%) and more disliked (20%) the idea than liked (16%) as seen in Table 7. This is concurrent with the participants' answers in the previous question and shows that consumers are very unsure when it comes to functional meat products. The reason for consumers having a less positive attitude towards bioactives in meat is likely influenced by "fear of the unknown" style response. Very little functional food meat based products exist within the European food market, and perhaps with more familiarity with these products consumers will begin to place more trust in them.

3.3.4 Consumer knowledge of Co-enzyme Q10 and acceptance within processed meats

Page 4 of the questionnaire quizzed participants' knowledge of CoQ10. A total of 44% of respondents claimed to have heard of CoQ10 the other 56% claimed no knowledge. Women were found to be divided directly in half with 50% answering they have heard of CoQ10 and 50% answering no (Table 8). Males were divided 30% saying yes and 70% for no (Table 8). Women we statistically (P<0.05) more likely to have heard of CoQ10 and men were statistically (P<0.05) more likely not to have heard of it (Table 8). Age of respondents also had a statistical significance (P<0.05) on knowledge of CoQ10 consumers aged 26-30 were more likely to have heard of CoQ10 than those aged 18-20 (Table 8), however, no other statistical differences were found between other age groups.

When asked to select what health benefits are associated with CoQ10 the most commonly known amongst respondents was heart health with 74% of those who answered yes to having heard of CoQ10 choosing it as an associated benefit (Table 8).

Anti-ageing was the second most associated benefit with CoQ10 at 70% (Table 8). Anti cancer benefit was chosen by 34% of people, the other benefits included; migraine, Parkinson's, diabetes and weight loss were chosen between 14 and 18% of the time by participants.

Over 80% of participants had no knowledge of a natural dietary source CoQ10, no significant values were found between age, gender and consumption rate. Of those who answered yes 75% said fish was a source, 68% said meat, 42% said fruit and vegetables and 3% said I don't know (Table 8).

When asked whether or not you would consume a processed meat product containing CoQ10 70% of consumers said yes they would (Table 9) and 30% said they would not. Consumers' acceptance of functional foods was found by Verbeke (2005) to be highly dependent on the belief of the consumer in the health effects of said food. Considering only 56% had never heard of CoQ10 and would not be aware of any health benefit this number is quite large. No significant differences were found between age and gender, however, those who consume processed meats at least once a week were statistically (P<0.05) more likely to say yes (Table 9). In terms of willingness to pay more for a processed meat product containing bioactive compound respondents more commonly answered no (60%) and only 40% said that they would be willing to do so. Some significant differences were found between levels of consumption (Table 9) but none were found between age and gender. Many studies have shown that consumer willingness to use functional food is multi-conditional (Verbeke, 2005; Cox, Koster & Russell, 2004; Urala & Lähteenmäki, 2004; Bech-Larsen & Grunert, 2003) and this present study shows that cost is a major consideration in the use of functional foods for consumers. There is further evidence which suggests that consumers are highly sceptical of health and nutrition claims on

packages viewing the claims as attempts by the manufacturer to sell more of their product (Levy & Stokes, 1987).

3.4 Conclusion

The majority of consumers' attitude towards processed meat is one of an unhealthy product. Most believe processed meats contain large quantities of harmful chemicals, fat and especially salt, however roughly 20% of consumers reported as being unsure for all but salt. However, the data obtained can be seen as skewed by the higher levels of women who responded than men. Women have been shown in previous studies to be more health conscious than men and if the study was conducted obtaining equal male responses the percentage of consumers who dislike processed meats are likely to decrease significantly. Even though consumer views on processed meats presently lean towards a negative one the processed meats industry is still a vital sector within the EU agricultural economy. Health officials and consumers alike must keep in mind that the processed meat industry turns low economical cuts of meat into in some cases highly valuable products which in themselves are a source of valuable protein and other nutrients including many bioactive compounds which provide health benefits. Many of these products also possess traditional status and are a source of local income and pride throughout numerous cities and towns in Europe.

Consumers were found to be very pro-bioactive compounds in yogurt style products but unsure of their feelings on the idea of them in meat based products, which is likely due to the lack of familiarity to these products. Many of the respondents were willing to consume meat based functional food products but were not willing to pay more for them. Despite all the known nutritional benefits of meat, innovation and science based product development remain extremely weak especially within the red meat sector of the meat industry (Troy & Kerry, 2010). The meat industry needs to paint a balanced picture to the consumer of the health and nutritional value of meat compared to the negative health issues associated with the consumption of meat. More investment is needed to portray processed meats as a not being a solely unhealthy product and that it can be used as a vehicle for functional ingredients. Salt reduction in foods in particular is highly important to consumers and is vital for processed meats to be seen in a good light by the European consumer. It is the opinion of this author that processed meats can if given the funding be seen in a more agreeable light by the European consumer and could over take dairy as the front runner in the functional food sector.

3.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

3.6 Table and Figures

1 abic 1. 1 age 2 survey questions	Table	1.	Page	2	survey	q	uestions
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Q4	Processed meat products are perfectly healthy to eat.
Q5	Processed meats are unhealthy to eat.
Q6	Processed meats are healthy as part of a larger balanced diet.
Q7	Processed meats are full of chemicals that may have negative effects to your health.
Q8	Processed meats contain a large quantity of fat.
Q9	Processed meats contain a large quantity of salt

Table 2. Page 3 survey questions

Q10	How do you feel about bioactive compounds such as plant sterols or probiotics?
Q11	How do you feel about bioactive compounds in dairy products such as yogurts?
Q12	How do you feel about bioactive compounds in processed meat products such as burgers?
Q13	Do you think processed meat products would be a suitable carrier for bioactive compounds?

Table 3. Page 4 Questions and Responses

Q14	Have you ev	er heard of Co	o-enzyme Q1	0?			
A	•Yes	•No					
Q15	Please select	t the health be	nefits you thi	ink are associa	ted with Co	-enzyme Q10	:
A	•Heart Health	•Migraine Relief	•Cancer	•Parkinson's Disease	•Diabetes	•Weight loss	•Anti-ageing
Q16	Do you knov	w any natural	dietary sourc	e of Co enzym	ne Q10?		
A	•Yes	•No					
Q17	Which of the	ese food group	os do you thin	nk naturally co	ontain Co-er	zyme Q10:	
A	•Fish	•Meat	•Fruit and Veg	•Don't Know			

Table 4. Page 5 Questions and Responses

Q18	Would you consume a processed meat product which contained Co-enzyme
A	•Yes •No
Q19	Would you be willing to pay more for a processed meat product that contained a health beneficial bioactive compound such as Co-enzyme Q10?
A	•Yes •No

Table 5.Respondent Demographic.

Demographic predictor variables and associated levels	n	%
<u>Age:</u>		
18-20	14	2.55
21-25	56	10.22
26-30	67	12.23
31-35	62	11.31
36-40	72	13.14
41-45	74	13.50
46-50	70	12.77
51-55	67	12.23
56-60	38	6.93
61+	28	5.11
<u>Gender:</u>		
Female	388	70.80
Male	160	29.20
How Often Respondant consumes Processed Meat Product:		
Daily	27	4.93
A few times a week	213	38.87
Once a week	167	30.47
Rarely	115	20.99
Never	26	4.74

_	Age %													Degree of Consumption %				
	Total	18-20	21-25	26-30	31-35	36-40	41-45	46-50	51-55	56-60	61+	Female	Male	Daily	Multiple/week	1/week	Rarely	Never
Processed meat pro	ducts a	are perfectly	healthy to	eat.														
Strongly Agree	1	0 a	0 a	1 ^a	0 a	0 a	3 a	2 a	0 a	0 a	4 ^a	1 ^a	1 ^a	12 ^a	0 b	1 ^b	1 ^b	0 ^{ab}
Agree	16	15 ^a	20 a	10 ^a	18 ^a	14 ^a	15 ^a	14 ^a	16 ^a	24 a	19 ^a	12 ^a	27 ^b	32 a	25 a	10 ^b	7 b	8 ^{ab}
Unsure	20	31 a	19 ^a	22 a	17 ^a	13 a	26 a	15 ^a	19 ^a	26 a	27 a	19 ^a	21 a	16 ^a	27 a	19 ^a	14 ^a	4 ^a
Disagree	48	38 ^{ab}	46 ^{ab}	54 ^{ab}	52 ab	62 ^b	45 ^{ab}	56 ab	46 ^{ab}	24 a	38 ^{ab}	51 ª	44 a	32 ^{ab}	42 ^b	62 a	49 ^{ab}	40 ^{ab}
Strongly Disagree	14	15 ^a	15 ^a	12 ^a	13 ^a	11 ^a	12 ^a	14 ^a	18 ^a	26 a	12 ^a	17 ^a	7 ^b	8 ^{ab}	7 ^b	9 ^b	30 ac	48 ^c
Processed meats ar	e unhe	althy to eat.																
Strongly Agree	11	8 a	9 a	9 a	15 ^a	7 a	9 a	15 ^a	15 ^a	16 ^a	8 a	14 ^a	6 ^b	4 ^{ab}	4 ^b	11 ^b	20 ac	40 °
Agree	46	46 ^a	50 a	51 a	48 a	56 a	45 a	55 a	43 a	26 a	38 a	51 ^a	38 ^b	36 ab	40 ^b	55 a	56 ab	32 ^{ab}
Unsure	22	38 a	20 a	24 a	17 ^a	24 ^a	23 a	15 ^a	18 ^a	34 a	31 a	21 a	26 a	24 a	28 ^a	22 a	15 ^a	12 ^a
Disagree	17	8 a	19 ^a	15 ^a	20 a	13 ^a	20 a	15 ^a	19 ^a	21 a	15 ^a	13 ^a	27 ^b	24 ab	27 ^b	11 a	9 a	8 ^{ab}
Strongly Disagree	2	0 a	2 a	1 ^a	0 a	0 a	3 a	0 a	4 a	3 a	8 a	2 a	3 a	12 ^a	1 ^b	1 ^b	1 ^b	8 ^{ab}
Processed meats ar	e healt	hy as part o	f a larger b	alanced d	iet.													
Strongly Agree	5	8 a	7 a	4 a	3 a	7 a	5 a	5 ^a	4 a	11 ^a	0 a	3 a	10 ^b	32 a	8 ^b	2 bc	0 c	0 bc
Agree	43	46 ^a	52 a	51 ^a	52 a	42 a	43 a	39 a	30 a	42 a	46 ^a	41 ^a	50 a	44 ^{ab}	52 b	49 ^b	3 ac	8 ^c
Unsure	18	31 a	15 ^a	18 ^a	15 ^a	20 a	26 a	14 ^a	21 a	16 ^a	15 ^a	18 ^a	20 a	12 ^a	22 a	18 a	16 ^a	12 ^a
Disagree	25	15 ^a	24 a	24 a	18 ^a	27 a	22 a	36 a	34 a	21 a	31 a	30 a	17 ^b	12 ^a	17 ^a	27 ^{ab}	43 ^b	36 ^{ab}
Strongly Disagree	6	0 a	2 a	3 a	12 ^a	4 a	4 a	6 ^a	10 ^a	11 ^a	8 a	7 a	3 a	0 ab	1 ^b	3 ^b	12 ^a	44 ^c
Processed meats ar	e full o	f chemicals	that may h	nave negat	ive effects	to your he	alth.											
Strongly Agree	9	8 a	6 a	1 ^a	12 ^a	7 a	8 a	14 ^a	9 a	16 ^a	12 ^a	11 ^a	3 ^b	4 ^{ab}	3 ^b	6 ^b	17 ^{ac}	40 ^c
Agree	41	46 ^a	43 a	42 a	32 a	45 a	45 a	42 ^a	51 a	32 a	42 a	45 a	35 ^b	24 ^{ab}	34 ^b	48 ^{ab}	51 ^a	52 ^{ab}
Unsure	29	15 ^a	30 a	34 a	28 a	30 a	30 a	27 a	22 a	39 a	38 a	28 a	33 a	44 a	37 a	26 ab	24 ^{ab}	4 ^b
Disagree	17	23 a	17 ^a	21 a	28 a	18 ^a	15 ^a	17 ^a	16 ^a	11 a	8 a	14 ^a	26 ^b	20 abcd	24 ^{cd}	20 bd	6 a	4 abcd
Strongly Disagree	2	8 a	6 a	1 a	0 a	0 a	3 a	0 a	1 ^a	3 a	0 a	1 ^a	4 ^b	8 a	2 a	1 a	1 ^a	0 a
Processed meats co	ntain a	a large quan	tity of fat.															
Strongly Agree	13	31 ^a	13 ^a	6 a	15 ^a	11 ^a	12 ^a	18 ^a	18 ^a	16 ^a	12 ^a	16 ^a	8 ^b	4 ^{ab}	7 ^b	13 ^{ab}	23 ac	44 ^c
Agree	54	62 a	56 a	57 a	53 a	59 a	53 a	56 a	51 a	53 a	58 a	54 ^a	58 a	48 ^{ab}	54 ^{ab}	63 ^b	51 ^{ab}	32 ^a
Unsure	19	8 a	22 a	22 a	20 a	21 a	22 a	14 ^a	19 ^a	18 ^a	19 ^a	18 ^a	23 a	32 a	23 a	15 a	17 ^a	20 ^a
Disagree	11	0 a	9 a	15 ^a	12 ^a	8 a	14 ^a	12 ^a	12 ^a	11 a	12 ^a	12 ^a	11 ^a	16 ^a	16 ^a	9 a	7 a	4 ^a
Strongly Disagree	0	0 a	0 a	0 a	0 a	0 a	0 a	0 a	0 a	3 a	0 a	0 a	0 a	0 a	0 a	0 a	1 ^a	0 ^a
Processed meats co	ntain a	a large quan	tity of salt.															
Strongly Agree	29	31 a	26 a	31 a	32 a	28 a	24 a	35 a	34 a	26 a	27 a	35 a	17 ^b	16 ^{ab}	22 ^b	31 abc	38 ac	56 °
Agree	58	69 ^a	63 ^a	55 a	53 a	65 a	64 ^a	61 ^a	52 a	68 a	54 a	58 a	65 a	60 ^a	64 ^a	63 a	52 a	40 ^a
Unsure	8	0 a	7 a	12 ^a	13 ^a	6 a	11 ^a	3 a	12 ^a	3 a	12 ^a	6 a	15 ^b	16 ^a	11 ^a	5 a	9 a	4 ^a
Disagree	2	0 a	4 ^a	1 ^a	2 a	1 ^a	1 ^a	2 a	1 ^a	3 a	8 a	2 ^a	3 a	8 ^a	3 a	1 ^a	1 ^a	0 ^a

Table 6. Questions on processed meats and frequency of answers as percentage.

^{abcd} within row and question, percentages with a common letter are not significantly different (P>0.05)

						Age	Gende	er %	Degree of Consumption %									
	Total	18-20	21-25	26-30	31-35	36-40	41-45	46-50	51-55	56-60	61+	Female	Male	Daily	Multiple/week	1/week	Rarely	Never
How do you feel	about	bioactive o	compound	s such as p	plant sterc	ls or prob	iotics?											
Strongly Like	8	0 a	9 a	12 ª	15 ^a	7 a	7 a	5 a	9 a	5 a	8 a	9 a	7 a	12 a	5 ^a	13 ^a	6 a	13 ^a
Like	48	25 a	46 a	46 a	45 ^a	56 a	42 a	62 a	53 a	57 a	46 a	50 a	50 a	40 a	50 ª	51 ^a	53 a	39 ^a
Unsure	32	58 a	31 a	38 a	30 a	31 a	40 a	25 a	32 a	30 a	38 a	33 a	34 a	32 a	37 a	330 a	32 a	26 ^a
Dislike	7	17 ^a	11 a	3 a	10 ^a	6 ^a	10 ^a	8 a	3 a	5 a	8 a	7 ^a	8 a	16 ^a	6 ^a	6 a	7 a	17 ^a
Stongly Dislike	1	0 a	2 a	0 a	0 a	0 a	1 ^a	2 a	3 a	3 a	0 a	1 ^a	1 a	0 a	1 a	11 ^a	2 a	4 ^a
How do you feel	about	bioactive o	compound	s in dairy p	oroducts su	uch as yo	gurts?											
Strongly Like	10	0 a	9 a	14 ^a	22 ^a	10 ^a	7 ^a	8 a	11 ^a	11 ^a	8 a	12 ^a	7 ^a	12 ^a	7 ^a	16 ^a	8 a	17 ^a
Like	52	50 a	52 a	51 ^a	45 ^a	56 a	56 a	60 a	64 a	57 a	50 a	55 ^a	53 a	44 a	55 ^a	54 ^a	61 ^a	35 ^a
Unsure	26	50 ^a	28 a	34 ^a	27 ^a	26 ^a	27 ^a	28 a	18 ^a	22 a	33 ^a	25 ^a	31 ^a	28 a	32 ^a	24 ^a	23 ^a	30 ^a
Dislike	6	0 a	9 a	2 a	5 a	9 a	10 ^a	5 a	6 a	5 a	8 a	6 ^a	7 a	16 ^a	5 a	6 a	6 a	9 ^a
Stongly Dislike	1	0 a	2 a	0 a	2 ^a	0 a	0 a	0 a	2 a	5 ^a	0 a	1 ^a	1 ^a	0 ^{ab}	0 b	0 ^b	2 ab	9 ^a
How do you feel	about I	bioactive o	compound	s in proces	sed meat	products	such as b	urgers?										
Strongly Like	2	0 a	6 a	3 a	2 a	1 a	0 a	2 a	8 a	0 a	0 a	2 a	3 a	8 a	3 a	2 a	1 a	4 ^a
Like	15	17 ^a	22 a	23 a	12 ^a	19 ^a	12 ^a	12 a	11 a	16 ^a	13 ^a	14 ^a	20 a	12 a	18 ^a	19 ^a	10 a	4 ^a
Unsure	59	58 a	54 a	60 ^a	68 a	57 a	70 a	66 a	61 ^a	54 ^a	58 a	63 ^a	58 a	48 a	67 ^a	61 ^a	62 a	43 ^a
Dislike	15	25 a	17 ^a	12 ^a	13 ^a	17 ^a	16 ^a	12 a	17 ^a	16 ^a	17 ^a	16 ^a	14 ^a	28 a	11 ^a	16 ^a	19 ^a	22 ^a
Stongly Dislike	4	0 a	2 a	2 a	5 ^a	6 ^a	0 a	8 a	5 a	14 ^a	13 ^a	5 ^a	5 a	4 ^{ab}	2 ^b	3 ^b	8 ab	26 ^a
Do you think pro	cessed	l meat pro	ducts wou	ld be a sui	table carri	er for bioa	active com	pounds?										
Strongly Like	3	0 a	4 a	3 a	2 a	3 a	1 ^a	3 a	8 a	0 a	0 a	2 ^a	4 a	4 a	4 ^a	3 a	1 ^a	4 ^a
Like	12	8 ^a	17 ^a	23 ^a	15 ^a	14 ^a	11 ^a	11 ^a	6 ^a	8 ^a	8 a	12 ^a	16 ^a	20 ^{ab}	17 ^b	14 ^{ab}	5 ^a	0 ^{ab}
Unsure	60	75 ^a	54 a	52 a	58 a	54 ^a	71 ^a	65 a	71 ^a	70 a	67 a	62 ^a	62 a	56 a	59 ^a	68 a	63 a	65 ^a
Dislike	16	17 ^a	17 ^a	18 ^a	18 ^a	21 ^a	16 ^a	12 ^a	14 ^a	11 ^a	21 ^a	18 ^a	13 ^a	8 a	18 ^a	11 ^a	22 a	22 ^a
Stongly Dislike	5	0 a	9 a	3 a	7 a	7 a	0 a	9 a	2 a	11 a	4 a	6 ^a	5 a	12 ^{ab}	2 ^b	5 ^{ab}	10 a	9 ^{ab}

Table 7. Questions on Bioactives and frequency of answers as percentage.

Stongly Dislike 5 0 a 9 a 3 a 7 a 7 a 0 a 9 a 2 a 11 a 4 a 6 a 5 a^{b} within row and question, percentages with a common letter are not significantly different (P>0.05)

			2		1	Âge	e %	Gender %			Degree of Consumption %							
	lotal	18-20	21-25	26-30	31-35	36-40	41-45	46-50	51-55	56-60	61+	Female	Male	Daily	Multiple/week	1/week	Rarely	Never
Have you ever heard	of Co-e	enzyme Q10	?															
Yes	44	0 a	43 ^{ab}	56 ^b	51 ^{ab}	41 ^{ab}	38 ^{ab}	46 ^{ab}	45 ^{ab}	50 ^{ab}	30 ab	50 ª	29 ^b	38 a	35 ª	50 a	50 a	59 ^a
No	56	100 a	57 ^{ab}	44 ^b	49 ^{ab}	59 ab	63 ^{ab}	54 ^{ab}	55 ^{ab}	50 ^{ab}	70 ab	50 ª	71 ^b	62 a	65 a	50 a	50 a	41 ^a
Please select the hea	lth ben	efits you thir	nk are ass	ociated wi	th Co-enzy	me												
Heart Health	74																	
Migrane	14																	
Cancer	34																	
Parkinsons Disease	18																	
Diabeties	17																	
Weight loss	17																	
Anti-Ageing	70																	
Do you know any natu	ural die	tary source	of Co enzy	/me Q10?														
Yes	83	0 a	25 a	26 ^a	15 ^a	12 ^a	18 ^a	15 ^a	15 ^a	11 ^a	9 a	17 ^a	15 ^a	21 ^a	11 ^a	18 ^a	23 a	14 ^a
No	17	100 ^a	75 a	74 ^a	85 a	88 ^a	82 a	85 a	85 a	89 ^a	91 ^a	83 ^a	85 ^a	79 ^a	89 a	82 a	77 ^a	86 ^a
Which of these food g	groups of	do you think	naturally	contain Co	o-enzyme													
Fish	75																	
Meat	68																	
Fruit and Veg	42																	
Don't Know	4																	

Table 8. Questions on Co-Enzyme Q10 and frequency of answers as percentage.

^{ab} within row and question, percentages with a common letter are not significantly different (P>0.05)

I				-		Age	%	Gende	r %		Degree of Consumption %							
	Total	18-20	21-25	26-30	31-35	36-40	41-45	46-50	51-55	56-60	61+	Female	Male	Daily	Multiple/week	1/week	Rarely	Never
Would y	ou cons	sume a pro	cessed m	ieat produ	ict which c	contained	Co-enzym	ne Q10?										
Yes	70	92 ^a	80 a	79 ^a	78 ^a	66 ^a	65 ^a	69 ^a	55 ^a	64 ^a	66 ^a	68 ^a	75 ^a	88 ^{ab}	88 ^b	76 ^a	38 ^c	0 d
No	30	8 ^a	20 a	21 ^a	22 a	34 ^a	35 ^a	31 ^a	45 ^a	36 a	34 ^a	32 ^a	25 ^a	12 ^{ab}	12 ^b	24 ^a	62 °	100 d
Would y	ou be w	villing to pa	iy more fo	r a proces	ssed meat	product th	hat contai	ned a hea	Ith benefic	cial bioacti	ve comp	ound such a	is Co-enz	yme Q10?				
Yes	40	42 ^a	24 a	40 a	39 a	36 a	42 a	52 a	34 ^a	39 a	48 a	41 ^a	35 a	29 abcd	43 ^{cd}	48 bd	24 a	23 abcd
No	60	58 ^a	76 ^a	60 a	61 ^a	64 ^a	58 a	48 a	66 ^a	61 ^a	52 a	59 ^a	65 ^a	71 abcd	57 ^{cd}	52 ^{bd}	76 ^a	77 abcd

Table 9. Questions on Co-Enzyme Q10 and frequency of answers as percentage.

^{abcd} within row and question, percentages with a common letter are not significantly different (P>0.05)

Chapter 4: Effect of varying salt and fat levels on the sensory quality of beef patties.

This Chapter is in the form of a manuscript accepted for publication in *Meat Science* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2012). Effect of varying salt and fat levels on the sensory quality of beef patties. *Meat Science*, 91, 460-465.

Abstract:

The interactive effects of varying levels of salt and fat on the sensory and physiochemical properties of beef patties were investigated. Twenty beef patties with varying levels of fat (30% 40% 50% 60% w/w) and salt (0.5%, 0.75% 1.0% 1.25% 1.5% w/w) were manufactured. All samples were assessed instrumentally for colour, moisture, fat, cooking loss and texture profile analysis. Sensory consumer evaluation was conducted using 25 consumers. The consumers rated each coded product, in duplicate, in terms of colour, texture, tenderness, juiciness, salt, taste, meat flavour, off flavour and overall acceptability. The data indicate that the most consumer acceptable beef patty was that containing 40% fat with a salt level of 1%. This is a 20% decrease in fat and a 50% decrease in salt levels when compared to commercial patties available in Ireland and the UK.

4.1 Introduction

Most research conducted on the development of meat products have empirically tested a limited number of ingredients, inclusion levels and process conditions on product yield and quality. This is a very logical and practical approach at one level, but does little to develop a deeper understanding of the processes occurring at the matrix level, all of which affect the final product. Implementation of a systematic scientific design approach in the controlled and efficient development of future meat products requires a basic application of chemical, biochemical, physical, biological and sensory principles and consideration of the meat system as a matrix of interacting components.

Myofibrillar proteins extracted during the mixing process denature and associate into a gel when the dispersion is heated (Foegeding & Lanier, 1987). This gel is a microsystem of a protein gel matrix containing an aqueous solution of salts and suspended particles. The texture of the processed meat product will depend on the structure of the matrix formed when the proteins gel, the amounts and types of particle and solutes entrapped in the gel matrix and the moisture content of the finished product (Foegeding & Lanier, 1987).

Recent studies have indicated that the intention to consume beef is gradually becoming more influenced by health and nutritional considerations (Fonseca & Salay, 2008; Angulo & Gil, 2007). The estimated cost of cardio vascular diseases to the EU economy has been estimated at €169 billion every year (Petersen, Peto, Rayner, Leal, Fernandez & Gray, 2005) while the estimated direct and indirect cost to the US from cardio vascular diseases in 2006 was \$403.1 billion (Thom, Hasse, Rosamond, Howard, Rumsfeld & Manolio, 2006). Currently, Irish and UK adults daily sodium intake is approximately three times the recommended daily allowance and therefore, public health and regulatory authorities are recommending reducing dietary intake of sodium to 2.4 g (6 g salt) per day (Desmond, 2006). Reports linking excessive sodium intake to the incidence of hypertension (Law, Frost & Wald, 1991a, 1991b; Dahl, 1972) is the main reason for reducing the sodium content of processed meats. A major portion of sodium in the diet is derived from processed foods, mostly in the form of sodium chloride (NaCl). Common table salt (NaCl) is used in the production of processed meat products primarily because of its functional capacity to solubilise myofibrillar proteins, thereby permitting efficient process-ability, all of which ultimately effect product texture, flavour and shelf life.

Consequently, salt reduction in meat products can have adverse effects on water and fat binding, impairing overall texture, increasing cooking loss and negatively impacting on sensory quality, especially taste. The perceived saltiness of NaCl is produced by the Na⁺cation in combination with the Cl⁻ anion (Miller & Bartoshuk, 1991).

High fat levels also present a greater risk of obesity and type 2 diabetes. The prevalence of obesity is increasing around the world and it is a significant public health problem in many countries (International Obesity TaskForce, 2002). Fats and oils play vital functional and sensory roles in various food products. Fats interact with other ingredients to develop texture, mouth feel and assist in the overall sensation of lubricity of foods (Giese, 1996). The proposed relationships between high cholesterol level and low polyunsaturated/saturated fatty acids (PUFA/SFA) ratio and the rise in coronary heart diseases has resulted in focusing on high fat food products including some meat products (Giese, 1992). Therefore, researchers have been working on strategies to reduce animal fat usage in meat products. The objective of this study was

to investigate the interactive effects of varying levels of salt in conjunction with varying levels of fat on the sensory and physio-chemical properties of beef patties.

4.2 Materials and Methods

4.2.1 Sample preparation

Beef was selected on the basis of a visual lean content of 95% and an ultimate pH of between 5.4 and 6.0. The beef was purchased along with beef back fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The beef and fat were then vacuum packed and storedl & until required for beef patty production. The frozen meat and fat was then cut into strips and allowed to thaw slightly before being minced through a mincer (Mincer Type: P14 TALSABELL S. A., Spain). The minced beef and fat was then blended together according to the formulations shown in Table 1. The respective experimental salt levels were then added and mixed thoroughly into the meat and fat by a Stephan mixer UMC5 (Stephan U. Sohner GmbH and Co., Germany) for 45 seconds (Table 1). The mix was then weighed into portions of 100 g and formed in to patties between grease proof papers using a patty press. The patties were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored at 4°C overnight.

4.2.2 Cooking

All samples were wrapped in foil and dry cooked at 150°C in a Zanussi convection oven (C. Batassi, Conegliano, Italy) for approximately 12 minutes to an internal temperature of 73°C, as measured by an internal temperature probe (Testo 110, Lenzkirch, Germany). All test samples were cooked at the same time and segregated to prevent any mixing.
4.2.3 Sensory evaluation

Sensory analysis was performed using 25 consumers in the age range 20–30 years. All were selected on the basis that they consume and purchase beef patties regularly. For each patty, consumers were asked to indicate their score on a 10 cm line scale ranging from 0 at the left to 10 at the right and rating subsequently scored in cm from the left. Consumers were asked to evaluate the patties using the following descriptors: colour, coarseness, toughness, juiciness, salt taste, meat flavour, off-flavour and overall acceptability. The off-flavour term was explained to consumers as off-flavour, rancid, cardboard or linseed oil-like flavour. Sensory analysis was undertaken in the panel booths at the university sensory laboratory that conforms to ISO (1988) international standard. Five coded samples were presented to the consumers and they were required to rinse with water before tasting each sample. Sample presentation order was randomised to prevent any flavour carryover effects (MacFie, Bratchell, Greenhoff & Vallis, 1989). All samples were presented in duplicate.

4.2.4 Protein content

Protein concentrations were measured using the Kjeldahl method (Suhre, Corrao, Glover & Malanoski, 1982). The digestion block was pre-heated to 410°C. Approximately 0.5 g of well homogenised sample was weighed accurately into a digestion tube. 15 ml of sulphuric acid (nitrogen free), 10 ml hydrogen peroxide and 2 "kjeltabs" were added to the sample. The tubes where then placed in the heated digestion block. Samples were removed from the block when the samples became colourless. After removal, the tubes were allowed to cool in the fume hood. Subsequently, 50 ml of distilled water was added carefully to the cooled and digested sample in the fume-hood. The tubes were placed in the distillation unit along with a receiver flask containing 50 ml of 4% Boric acid with indicator. On completion, the contents of the receiver flask were titrated against 0.1 N hydrochloric acid until the green colour reverted back to the original red colour.

4.2.5 Ash content

Ash content was determined by muffle furnace (AOAC, 1923). A muffle furnace was pre-heated to 525°C. Approximately 5 g of well blended sample were weighed into porcelain dishes using a balance that weighs to 1 mg. The dishes containing samples were then placed in the muffle furnace for (approximately 6 hours) until the samples went white in colour. The dishes were then removed and placed in a desiccator to cool. The dishes were then weighed and the ash content calculated.

4.2.6 Moisture and fat content

A 200 g sample was homogenised using Büchi Mixer B-400 and quickly transferred into a moisture proof bag to avoid loss. Moisture and fat content were then determined using the CEM SMART (moisture) and SMART Trac (fat) systems (Bostian, Fish, Webb, & Arey, 1985).

4.2.7 Colour

The surface colour of cooked and raw patties was measured according to the CIE L* a* b* colour system using a Minolta CR 300 colorimeter (Minolta Camera Co. Ltd., Osaka, Japan) with an 11 mm-diameter aperture, D65 illuminant, calibrated by the CIE Lab colour space system using a white tile (C: Y=93.6, x=0.3130, y=0.3193), (Minolta calibration plate). Colour measurements (CIE L*, a* and b* values representing lightness, redness and yellowness, respectively) were taken before cooking and immediately after the cooked samples were cooled to room temperature. Nine readings were taken per sample.

4.2.8 Cooking loss

Beef patties sample weights were recorded before and after cooking and the differences in weights recorded. Beef patties were wrapped in foil and cooked in a Zanussi convection oven at 150°C for 10 minutes to reach an internal temperature greater than 72°C. Before weighing, samples were blotted with a paper towel to remove excess surface moisture.

Calculation for cook loss was as follows:

% cook loss = ((cooked weight - raw weight)/ raw weight) x 100

4.2.9 Texture analysis

Texture measurements in the form of texture profile analysis (TPA) were performed at room temperature with a Texture Analyser 16 TA-XT2i (Stable Micro Systems, Surrey, UK). Three patties were taken and subjected to a two-cycle compression test using the 25 kg load cell. The samples were compressed to 40% of their original height with a cylindrical probe (SMSP/100 Compression plate) 100 mm diameter and a cross-head speed of 1.5 mm/s. Mean values were expressed in terms of peak force (KgF). Texture profile parameters were determined following descriptions by Bourne (1978) and the SMS manual (Stable Micro Systems, Surrey, UK). All analyses were performed in triplicate.

4.2.10 Data analysis

ANOVA-partial least squares regression (APLSR) was used to process the raw data accumulated from the 25 test subjects during the sensory evaluation and data acquired by instrumental methods. The X-matrix was designed as 0/1 design variables for fat and salt content of patties. The Y-matrix was designed as sensory, chemical, and instrumental variables. The optimal number of components in the APLSR models presented was determined to be four principal components (Fig. 1). PC 1 versus PC 3 is presented; the other PCs did not yield additional information or provide any predictive improvement in the Y-matrix obtained through their examination. The validated explained variance for the model constructed was 35.77% and the calibrated variance was 36.37%. To derive significance indicators for the relationships determined in the quantitative APLSR, regression coefficients were analysed by Jack-knifing (Table 2) which is based on cross-validation and stability plots (Martens & Martens, 1999, 2001). All analyses were performed using the Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway).

4.3 Results and Discussion

4.3.1 Sensory consumer evaluation

Sensory evaluation and instrumental data are presented in the APLSR plot in Figure 1 with the corresponding ANOVA values in Table 2 for fat and Table 3 for salt effects. From Figure 1, in the upper left hand quadrant red colour (Colour) can be seen to be correlated with the lower fat patties. Also from Table 2, colour was significantly (Pb0.001) negatively correlated to the 40% fat beef patties and (P<0.001) positive for the 60% fat patties, which can be explained by the increased fat levels in the latter.

The 0.5% salt containing patties were significantly (P<0.01) correlated to colour, whereas the other salt levels were not, this is likely linked to the higher salt patties having more bound moisture compared to the low salt patty. Ventanas, Puolanne, & Tuorila (2010) found that sausages with higher salt content had a more intense colour and were perceived to be shinier than lower salt sausages. From Figure 1, coarseness (texture), toughness and juiciness were not clearly correlated to any specific product, however, by examining the ANOVA values from Table 2, significant correlations between these attributes and products were observed. Patties containing lower fat and less salt tended to have a less coarse mouth-feel when compared to higher fat and higher salt patties. However, these figures were not significantly different. Lower fat patties and low salt patties were found to be significantly (P<0.05) more tender than higher fat and higher salt patties. The 30% and 40% fat patties were shown to be negatively correlated (P<0.05) to toughness, whereas 50% and 60% fat patties were shown to be positively correlated to toughness, but not in a significant manner. One explanation for this may be the higher fat patties had more cook-out which resulted in them being perceived as tougher by assessors.

The 1.5% salt patties were found to be significantly (P<0.05) correlated to toughness, whereas, the 1.25% salt patties were also positively correlated to toughness, but this correlation was not significant. The 0.75% salt patties were shown to be significantly (P<0.05) negatively correlated to toughness along with the 1% and 0.5% salt patties, which were also found to be negatively correlated to toughness, but not in a significant manner. The 30% fat patties were significantly (P<0.05) and negatively correlated to toughness and the 40% fat patties were significantly (P<0.05) and negatively correlated to toughness to a greater extent. Therefore the most tender products were shown to be those that contained 0.75% salt and 40% fat patties. The tenderness value seems to be highly dependent on the salt to lean meat ratio which is lowest in the low salt and low fat patties. The reduction in salt means a decrease in the solubilisation of myofibrillar protein which in turn has been shown to decrease toughness (Schwartz & Mandigo, 1976). From Figure 1 the level of fat appears to have had no significant effect on sensory juiciness, however by examining Table 2, a trend can be observed showing that higher fat patties are directionally positively correlated to higher juiciness. These results are in partial agreement with previous research carried out by Berry & Leddy (1984) who found that ratings for juiciness in higher fat patties were higher than that of lower fat patties. However, Kenddall, Harrison & Dayton (1974) showed no effects of fat level on juiciness scores for ground beef patties.

Salt levels had a much more significant effect on juiciness with the 1.25% and 1.5% salt patties which were significantly (P<0.01) juicier compared to the 0.5%, 0.25% and 0.75% salt patties which were significantly (P<0.05) less juicy. This is due to the water holding capacity of the myofibrillar proteins which are solubilised by the salt. More salt means increased solubilisation which in turn increases the water

holding capacity and the perceived juiciness by the consumer. The perception of salt taste from Fig. 1 and Table 2 can clearly be seen to be highly correlated with the higher salt containing patties, evident in the upper left hand quadrant of the APLSR plot. The ANOVA values from Table 2 show a strong positive and significant correlation (P<0.001) for the 1.5% and 1.25% patties to salt taste and a strong negative and significant correlation (Pb0.001) for the 0.75% and 0.50% patties to salt taste.

The ANOVA values from Table 2 also show that salt perception was negatively significant for 30% (P<0.05) and the 40% (P<0.01) fat beef patties, respectively (Table 1). The 50% fat beef patties were also significantly (P<0.001) and positively correlated to salt taste. Similar effects have also been demonstrated by Matulis, McKeith, Sutherland & Brewer (1995) and Ruusunen et al. (2005) who found that the effect of meat content on perceived saltiness in Frankfurters, as well as beef and pork meat patties, was stronger than the effect of fat content. Salt levels had the greatest effect on salt perception with significant (P<0.001) values for 1.5% and 1.25% having increased saltiness and 0.75% and 0.5% salt showing low saltiness values. Meat flavour was not significantly affected by any of the salt levels (Table 2). However, the 40% fat beef patties were positively significant (P<0.01) for meat flavour (Fig. 1, Table 2). Higher fat levels were significantly (P<0.05) less meaty than lower fat levels. Off-flavour correlated with patties containing 50% and 60% fat and was significantly (P<0.001) higher for 60% fat and significantly lower (P<0.001) for 40% fat containing patties. The higher fat level in the 60% fat beef patties increased the propensity for lipid oxidation which is likely displaying the correlation to offflavour, similar findings have been shown by (Morrissey, Sheehy, Galvin, Kerry & Buckley, 1998; Faustman & Cassens, 1989).

From Table 2 the 0.5% salt patties were (P<0.05) positively significant for Overall Acceptability whereas the other salt levels showed no significant difference. Fat levels showed a significantly (P<0.05) and (P<0.01) negative correlation to Overall Acceptability for 50% and 60% fat levels respectively. Whereas the 40% fat patties were positively significant (P<0.001) for acceptability.

4.3.2 Physiochemical analysis

From Figure 1 and Table 3 moisture content correlated with lower fat containing patties and higher salt containing patties, where moisture content of the cooked lower fat patties had significantly (P<0.001) higher moisture content than the higher fat patties. Similarly, fat content was significantly higher (P<0.001) in higher fat patties. However, cooking loss (Table 3) was significantly (P<0.001) affected by fat content, with 30%, 40% and 50% fat patties having decreased cooking losses and 60% fat patties having increased cooking loss, which is also displayed in Figure 1. The level of salt impacted on fat and moisture levels, with the higher salt patties (1.5%, 1.25% and 1.0%) correlating to significantly (P<0.001) less fat and a significantly (P<0.001) higher moisture content than the 0.75% and 0.5% salt patties.

Cooking loss was found to be significantly higher (P<0.01) from patties containing 0.75% salt and significantly lower (P<0.001) for patties containing 1.5% salt. The effect of increased salt levels on reducing cooking losses from meat patties was also demonstrated by Ruusunen et al. (2005). They showed that increased salt content reduced cooking losses for pork and beef patties.

Colour measurements showed that CIE L values were significantly (P<0.001) negatively correlated to both cooked and raw patties containing 30%, 40% and 50% fat, indicating that they were darker. From Figure 1 it is clearly shown that cooked

and especially raw L values are correlated to 60% fat, indicating that 60% fat patties are lighter in colour. The degree of redness, which is measured by the a-value in both cooked and raw samples is significantly (P<0.001) correlated to 30% and 40% fat patties and negatively correlated (P<0.001) to 60% fat (Figure 1). This is due to the higher proportion of red meat in the lower fat patties compared to the higher fat patties.

A correlation between hardness and cohesiveness to low fat patties in quadrant 1 and 2 is shown in Fig. 1. Results from Table 3 show that the texture analysis (hardness and cohesiveness) on cooked beef patties were significantly (P<0.001) correlated to patties containing 30%, 40% and 50% fat, whereas patties containing 60% fat were significantly (P<0.001) negatively correlated to hardness and cohesiveness. Patties containing 60% fat were found to be significantly (P<0.001) more springy and resilient when compared to patties containing all other fat levels. The decrease in hardness for patties containing 60% fat may be attributed to a decrease in myofibrillar proteins which form the gel network when extracted using salt (Acton & Dick, 1984) fat also has a natural lubricating effect in meat products.

Hardness and chewiness were significantly correlated (P<0.001) with the patties containing 1.5%, 1.25% and 1.0% salt while the lower salt patties 0.5% and 0.75% were more correlated to resilience (P<0.001) and springiness (P<0.05). The correlation of higher salt levels in patties to hardness can also be seen in quadrants 1 and 2 in Figure 1. The increase in salt levels causes more solubilisation of the functional myofibrillar proteins, thus forming a tougher product resulting in an increase in hardness and chewiness. The increase in hardness, in turn, increases the likeliness of fracturability which decreases the springiness and resilience of the product (Acton & Dick, 1984).

4.4 Conclusions

Fat and salt content have been shown to have a major effect on a number of quality attributes of beef patties. Consumer sensory data have shown that lower fat patties manufactured in this study were perceived to be darker in appearance and low salt patties were shown to be more tender, whereas Off flavour can be seen to be correlated to high fat patties. Also, cooking loss is shown to be most associated to higher fat patties and lower salt patties. Higher fat and lower salt patties were shown to be highly correlated to resilience and springiness.

Data from consumer sensory analysis indicate that the most acceptable patty formulation is 40% fat and 0.5% salt. This is a 20% decrease in fat and a 50% decrease in salt levels of average commercial patties. These patties can be characterised as being darker in colour, having less coarse mouth feel, being more tender, less juicy, less salty, meat flavour was not significantly affected, and these patties had less of an off flavour when compared to higher salt and higher fat patties.

4.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physicochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

4.6 Tables and Figures Table 1. Beef burger composition with respect to beef, fat and salt content.

	Composition				Composition			
Code	% Beef	% Fat	% Salt	Code	% Beef	% Fat	% Salt	
30F1.5S	68.50	30.00	1.50	50F1.5S	48.50	50.00	1.50	
30F1.25S	68.75	30.00	1.25	50F1.25S	48.75	50.00	1.25	
30F1S	69.00	30.00	1.00	50F1S	49.00	50.00	1.00	
30F0.75S	69.25	30.00	0.75	50F0.75S	49.25	50.00	0.75	
30F0.5S	69.50	30.00	0.50	50F0.5S	49.50	50.00	0.50	
40F1.5S	58.50	40.00	1.50	60F1.5S	38.50	60.00	1.50	
40F1.25S	58.75	40.00	1.25	60F1.25S	38.75	60.00	1.25	
40F1S	59.00	40.00	1.00	60F1S	39.00	60.00	1.00	
40F0.75S	59.25	40.00	0.75	60F0.75S	39.25	60.00	0.75	
40F0.5S	59.50	40.00	0.50	60F0.5S	39.50	60.00	0.50	

Table 2. Significance of estimated regression coefficients (ANOVA values) for the relationships between sensory terms ^a and instrumental measurements ^b as derived by Jack-knife uncertainty testing for various patty formulations containing varying levels of fat.

Attribute	30% Fat	40% Fat	50% Fat	60% Fat
Colour ^a	0.4913 [°] ns	0.0001***	-0.0029**	-0.0001***
Texture ^a	-0.7575 ns	-0.3900 ns	0.4997 ns	0.3891 ns
Toughness ^a	-0.0409*	-0.0144*	0.0042**	0.0558 ns
Juiciness ^a	-0.5521 ns	-0.1149 ns	0.9276 ns	0.0759 ns
Salt Taste ^a	-0.0337*	-0.0017**	0.0001***	0.6901 ns
Meat Flavour ^a	0.4918 ns	0.0003**	-0.0064**	-0.0109*
Off Flavour ^a	-0.1430 ns	-0.0001***	0.0007**	0.0001***
Acceptability ^a	0.5919 ns	0.0001***	-0.0180*	-0.0008**
Cooking Loss ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***
Cooked L Value ^b	-0.0001***	-0.0001***	-0.0001***	-0.8659 ns
Cooked a Value ^b	0.0001***	0.0001***	0.0035**	-0.0001***
Cooked b Value ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***
Raw L Value ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***
Raw a Value ^b	0.0001***	0.0001***	0.0429*	-0.0001***
Raw b Value ^b	0.0001***	0.0001***	0.0001***	-0.0001***
Moisture Content ^b	0.0001***	0.0001***	0.9565 ns	-0.0001***
Fat Content ^b	-0.0001***	-0.0001***	-0.0597 ns	0.0001***
Hardness ^b	0.0001***	0.0001***	0.0001***	-0.0001***
Springiness ^b	-0.0001***	-0.2774 ns	-0.0191*	0.0001***
Cohesiveness ^b	0.0001***	0.0001***	0.0001***	-0.0001***
Chewiness ^b	0.4563 ns	0.0001***	0.0001***	-0.9401 ns
Resilience ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c Estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates weather the correlation is positively or negatively correlated.

^d Significance of regression coefficients; ns, not significant; *P < 0.05, **P < 0.01, and ***P < 0.001.

Table 3. Significance of estimated regression coefficients (ANOVA values) for the relationships of sensory terms ^a and instrumental measurements ^b as derived by Jack-knife uncertainty testing for various patty formulations containing varying levels of salt.

Attribute	1.5% Salt	1.25% Salt	1% Salt	0.75% Salt	0.5% Salt
Colour ^a	-0.2313 ns	-0.1256 ns	0.2568 ns	0.3127 ns	0.0072**
Texture ^a	0.8249 ns	0.6837 ns	-0.4420 ns	-0.7823 ns	-0.6493 ns
Toughness ^a	0.0102*	0.1243 ns	-0.1737 ns	-0.0354*	-0.0926 ns
Juiciness ^a	0.0040**	0.0179*	-0.6708 ns	-0.0131*	-0.0017**
Salt Taste ^a	0.0001***	0.0001***	-0.4372 ns	-0.0001***	-0.0001***
Meat Flavour ^a	-0.5581 ns	-0.2888 ns	0.26989 ns	0.4886 ns	0.1519 ns
Off Flavour ^a	0.1056 ns	0.8029 ns	-0.1679 ns	-0.3578 ns	-0.8631 ns
Acceptability ^a	-0.2482 ns	-0.1543ns	0.2759 ns	0.2510 ns	0.0429*
Cooking Loss ^b	-0.0001***	-0.7563 ns	-0.7746 ns	0.0063**	0.3619 ns
Cooked L Value ^b	-0.0001***	-0.0002**	-0.0001***	0.0002**	0.0001***
Cooked a Value ^b	0.0031**	0.4624ns	0.4738 ns	-0.0009**	-0.3296 ns
Cooked b Value ^b	-0.0001***	-0.0205*	-0.0006**	0.1026 ns	0.0001***
Raw L Value ^b	-0.4838 ns	-0.2407 ns	-0.0041**	0.1231 ns	0.0001***
Raw a Value ^b	0.8984 ns	0.2122 ns	0.0093**	-0.3574 ns	-0.0001***
Raw b Value ^b	0.0004**	0.0745 ns	0.0175*	-0.2246 ns	-0.0001***
Moisture Content ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***
Fat Content ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***	0.0001***
Hardness ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***
Springiness ^b	-0.7746 ns	-0.7098 ns	-0.0845 ns	0.0757 ns	0.0090**
Cohesiveness ^b	0.0002**	0.1071 ns	0.6700 ns	-0.0001***	-0.4152 ns
Chewiness ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***
Resilience ^b	-0.0001***	-0.0001***	-0.0267*	0.0001***	0.0001***

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c Estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates weather the correlation is positively or negatively correlated.

^d Significance of regression coefficients; ns, not significant; *P < 0.05, **P < 0.01, and ***P < 0.001.

Figure. 1. APLSR for the various beef patty formulations.



▲=Percentage of fat or salt present in the beef patties, •=sensory descriptor, chemical and instrumental variables. The concentric circles represent 100% and 50% explained variance, respectively.

Chapter 5: The Impact of Salt and Fat Level Variation on the Physiochemical Properties and Sensory Quality of Pork Breakfast Sausages.

This Chapter is in the form of a manuscript accepted for publication in *Meat Science* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). The impact of salt and fat level variation on the physiochemical properties and sensory quality of pork breakfast sausages. *Meat Science*, 93, 145-152.)

Abstract:

The sensory and physiochemical properties of sausages with varying fat and salt levels were investigated. Twenty eight sausages were produced with varying concentrations of fat (22.5 % 27.5 % 32.5 % 37.5 % w/w) and salt (0.8 %, 1 %, 1.2 %, 1.4 %, 1.6 %, 2 %, 2.4 % w/w). Sausages were assessed instrumentally for colour, moisture, fat, cooking loss and texture profile analysis. Consumers (n = 25) evaluated each product in duplicate for colour, texture, tenderness, juiciness, salt taste, meat flavour, off-flavour and overall acceptability using a hedonic scale.

Lowering fat produced products which consumers rated as less dark in colour, tougher, less juicy and taste less salty than higher fat products. However, no significant preferred sample was found amongst consumers. Salt reduction in products produced sausages which consumers rated as paler in colour, more tender and with greater meat flavour than higher salt containing products. The sausages containing 1.4 % and 1.0 % salt were significantly (P < 0.01) found to be more acceptable to consumers than other salt levels.

5.1 Introduction

Studies in meat consumption in the last decade have shown that the health and nutritional value of a product is a major factor in consumer preference (Fonseca & Salay, 2008; Angulo & Gil, 2007). Cardiovascular disease (CVD) accounts for 30 % of all deaths across the world (World Health Organization, 2009). Hypertension, a term which describes high blood pressure, has high global prevalence. Many studies have shown a link between a high intake of dietary sodium and hypertension (Law, Frost & Wald, 1991a, 1991b; Dahl, 1972). The main source of sodium (75 % of total dietary intake) in most of our diets has been shown by Appel & Anderson (2010) to come from processed food. Processed meats can also contain high levels of animal fat. High levels of fat have been associated with increased risk of promoting obesity, diabetes and also cancers especially colon cancers (Aggett, Antoine, Asp, Bellisle, Contor & Cummings, 2005).

Even though salt and fats are shown to impact negatively on health they are still integral parts of any meat product. Salt is a vital ingredient in processed meat as it has numerous technological benefits such as preservation, taste enhancement and water binding (Durack, Alonso-Gomez & Wilkinson, 2008). Water holding capacity is defined as the ability of a food to enclose liquid within a three dimensional structure (Chantrapornchai & McClements, 2002). Salt is able to increase the water holding capacity of a meat product by extracting myofibrillar proteins which associate into a gel when heated (Foegeding & Lanier, 1987).

Fat also greatly contributes to the eating quality of meat (Webb, 2006; Wood, 1990). It can interact with other components present within a meat system and help to develop what can be a more consumer acceptable product. Affecting things like texture, mouthfeel and providing lubrication, as well as contributing to overall flavour

(Javidipour, Vural, Ozbas & Tekin, 2005; Crehan, Troy & Buckley, 2000; Wood, Enser, Fisher, Nute, Richardson & Sheard, 1999; Giese, 1996).

Reduced fat foods are seen by consumers to have inferior sensory properties than regular fat products and maintain a level of scepticism that there is a need for substitutes and additives used to replace fat. It can be argued that there is a great deal more to reduced fat products than just sensory acceptance (Hamilton, Knox, Hill & Parr, 2000). For instance, Levy & Stokes (1987) have shown evidence which also suggests that consumers are mistrustful towards product health claims, believing companies simply use these health benefit claims as a ploy to increase product sales. However, it is still important to obtain an acceptable limit at which salt and fat can be reduced from processed meat products without negatively impacting functionality, product quality or adversely affecting sensorial acceptability, so as to enhance the health status of processed meats. Work carried out by Tobin, O'Sullivan, Hamill & Kerry (2012a) and Tobin, O'Sullivan, Hamill & Kerry (2012b) have shown that fat and salt reduction can be successfully reduced in processed meat products such as burgers and Frankfurters.

This study aims to investigate the interactions between different salt and fat levels on the overall quality of cooked breakfast sausages and also to investigate the consumer optimisation of reduced salt and fat variants, without using fat and salt alternatives.

5.2 Materials and Methods:

5.2.1 Sample Preparation

Pork was selected on the basis of a high visual lean (V/L) score; pork shoulder was used with a V/L score of 99 % Pork was purchased along with pork back fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The meat and fat were vacuum packed and stored at -18 °C until required for sausage production. The frozen meat and fat were then cut into strips and allowed to thaw slightly before being minced through a 5 mm plate (TALSABELL S. A., Spain). The meat was weighed according to the formulations shown in Table 1 and fed into the bowl chopper. The required salt, seasoning and half the water was added and mixed at high speed for 60 seconds. The required fat was then added and the mix was chopped for further 60 seconds at high speed. The remaining water and rusk were then added and mixed at low speed for 15 seconds and high speed for 30 seconds. The sausage mix was then put into the casing filler and fed into collagen casings. The sausages were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored in chill over night at -4°C.

5.2.2 Cooking

Oven cooking was chosen as it was the most easily repeatable and controllable cooking method. All samples were wrapped in foil and dry cooked at 150 °C in a zanussi convection oven (C. Batassi, Conegliano, Italy) for 15 minutes to an internal temperature of 73 °C, as measured by an internal temperature probe (testo 110, Lenzkirch, Germany). All test samples were cooked at the same time to assure uniformity and segregated to prevent mixing.

5.2.3 Sensory Evaluation

Sensory analysis was carried out using 25 consumers within the age range of 20–30 years, following the method of O'Sullivan, Byrne & Martens (2003). Panellists were chosen on the basis that they regularly consume and purchase sausage style meat products. Sensory analysis was undertaken in the panel booths at the university sensory laboratory that conforms to ISO (1988) international standard. Five samples were presented to the consumers and they were required to rinse with water before tasting each sample. Sample presentation order was randomised to prevent any flavour carryover effects (MacFie, Bratchell, Greenhoff & Vallis, 1989). Consumers were asked to indicate their score on a 10 cm line scale ranging from 0 at the left to 10 at the right and rating was subsequently scored in cm from the left for each sausage presented. Consumers were required to evaluate the sausages using the following descriptors: colour, coarseness, toughness, juiciness, salt taste, meat flavour, off-flavour and overall acceptability. Off-flavour was described to consumers as off-flavour, rancid, cardboard or linseed oil-like flavour.

5.2.4 Protein Content

The Kjeldahl method (Suhre, Corrao, Glover & Malanoski, 1982) was used to measure protein concentrations. The digestion block was pre-heated to 410 °C. Approximately 0.5 g of well homogenised sample was weighed accurately into a digestion tube. 15 ml of sulphuric acid (nitrogen free), 10ml hydrogen peroxide and 2 "kjeltabs" were added to the sample. The tubes where then inserted in the heated digestion block. When the samples became colourless they were removed from the block. The tubes were allowed to cool in the fume hood after removal.

50 ml of distilled water was carefully added to the cooled and digested sample inside the fume-hood. The tubes and a receiver flask containing 50 ml of 4 % Boric acid with indicator were then placed into the distillation unit. After the sample had been distilled the contents of the receiver flask was titrated against 0.1 N hydrochloric acid until the green colour reverted back to the original red colour.

5.2.5 Ash Content

Ash content was determined using a muffle furnace (AOAC, 1923). A muffle furnace was pre-heated to 525°C. Approximately 5 g of well homogenised sample was weighed into porcelain dishes using a balance that weighs to 1 mg. The dishes containing samples were then put in the muffle furnace for approximately 6 hours until the colour of the samples went white. The dishes containing the samples were then removed and placed in a desiccator to cool. The dishes were then weighed and the ash content calculated.

5.2.6 Moisture and Fat Content

A Büchi Mixer B-400 (BÜCHI Labortechnik AG, Meierseggstrasse 40, Postfach, CH-9230 Flawil 1, Switzerland) was used to homogenise a total of 200 g of sausage sample. To avoid moisture or evaporative loss the homogenised sample was then quickly transferred into a moisture proof bag. Moisture and fat content were then determined using the CEM SMART (moisture) and SMART Trac (fat) systems (Bostian, Fish, Webb & Arey, 1985).

5.2.7 Colour

Both raw and cooked sausages were cut down the centre and were measured for colour according to the CIE L* a* b* colour system. Cooked samples were cooled to room temperature before measuring. A Minolta CR 300 colorimeter (Minolta Camera Co. Ltd., Osaka, Japan) with an 11 mm-diameter aperture and D₆₅ illuminant, calibrated by the CIE Lab colour space system using a white tile (C: Y = 93.6, x = 0.3130, y = 0.3193), (Minolta calibration plate) was used to conduct analysis. Colour measurements (CIE L*, a* and b* values representing lightness, redness and yellowness, respectively) were taken. Nine readings were taken per sample.

5.2.8 Cooking Loss

Sausage sample weights were recorded before and after cooking and the differences in weights were recorded. The sausages were wrapped in foil and placed in a Zanussi convection oven and cooked at 150 °C until an internal temperature of 73 °C tested by a temperature probe (Testo 110, Lenzkirch, Germany) was reached. Before weighing, samples were blotted with a paper towel to remove excess surface moisture.

Calculation for cook loss was as follows:

% cook loss = ((cooked weight – raw weight)/raw weight) x 100

5.2.9 Texture Analysis

Texture measurements were obtained for individual samples using texture profile analysis (TPA). All analysis were performed at room temperature with a Texture Analyser 16 TA-XT2i (Stable Micro Systems, Surrey, UK). Three individual (10 mm x 10 mm) cylindrical slices of sausage were taken from each batch. Every slice was then subjected to a two-cycle compression test using the 25 kg load cell. Samples were compressed to 40 % of their original height with a cylindrical probe (SMSP/35 Compression plate) 35 mm diameter and a cross-head speed of 1.5 mm/s.

Texture profile parameters were determined following descriptions by Bourne (1978) and the SMS manual (Stable Micro Systems, Surrey, UK). These included; hardness (N) maximum force required for the initial compression of the sample; springiness (mm), the ability of the sample to recover its original shape after the initial compression and the deforming force was removed; adhesiveness ($N \times mm$), area under the abscissa post initial compression; cohesiveness (dimensionless), extent to which the sample could be deformed prior to rupture, measured by the areas under the compression portion only and excludes the areas under the decompression portion instead of using the total area under positive force; chewiness ($N \times mm$), the required work to masticate the sample, measured as the product of hardness times cohesiveness times springiness; and resilience (dimensionless), the ratio between the negative force input to positive force input during the first compression. All analyses were performed in triplicate.

5.2.10 Data Analysis

The data accumulated from the 25 test subjects during the sensory evaluation and data acquired by instrumental methods were processed using ANOVA-partial least squares regression (APLSR). For Figure 1 the X-matrix was designed as 0/1design variables for fat and salt content of sausages, and as individual products for Figure 2. The sensory, chemical, and instrumental variables were used for the Ymatrix design. The optimal number of components in the APLSR models presented was determined to be four principal components; PC 1 versus PC 2 is presented for both Figure 1 and Figure 2; the other PCs did not yield any other additional information or provide any predictive improvement in the Y-matrix obtained prior to examination. The validated explained variance for the model constructed was 35.77% and the calibrated variance was 36.37% for Figure 1, and 32.73% validated explained variance and 33.40% calibrated variance for Figure 2. Regression coefficients were analysed by Jack-knifing (Table 4 and Table 5) to derive significance indicators for the relationships determined in the quantitative APLSR, which is based on crossvalidation and stability plots (Martens & Martens, 1999, 2001). All analyses were performed using the Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway).

5.3 Results and Discussion:

5.3.1 Sensory Consumer Evaluation

The results of the Consumer sensory evaluation which was carried out by 25 regular consumers of pork sausage and the instrumental data figures for percent fat and percent salt are presented in the APLSR plot in Figure 1 with the corresponding P values of the regression co-efficient in Table 4 for fat and Table 5 for salt effects.

Individual product correlations to sensory and physiochemical results are displayed in Figure 2.

Colour which was described as extremely pale to extremely dark was significantly correlated to samples containing higher fat which is shown in Figure 1 where colour and 37.5 and 32.5% fat samples are on the right hand hemisphere, and also in Table 4 where both higher fat levels are positively (P>0.001) correlated to colour. No salt levels are seen to be significantly correlated to colour in Figure 1 and Table 5, however low salt sausages are shown to be significantly negatively correlated to colour; this is similar to studies by Ventanas, Puolanne & Tuorila (2010) on bologna type sausages, where consumers perceived more intense colour in higher salt sausages.

Coarseness was found to be more correlated to lower fat with significant negative correlation to higher fat sausages (P>0.05) for 32.5% and (P>0.01) for 37.5% fat levels shown in Table 4. This is likely due to the lubricating effect of fat within a meat system (Javidipour et al. 2005; Crehan et al. 2000; Giese, 1996). Figure 1, clearly shows a correlation between lower salt levels and coarseness in its left hemisphere, this coincides with Table 5 where both were significantly negative (P>0.001) for 2% salt and significantly positive (P>0.001) for 1.2% salt correlations and indicate that lowering salt level increases the coarse mouthfeel of sausages to consumers. Similar findings were found by Tobin et al. (2012b) in Frankfurters wherein, when salt was reduced by 50% from 3% to 1.5% consumers measured a significant increase of coarseness in the samples presented. The reduction in salt is likely affecting the water and fat binding of the product, thus impairing the mouthfeel.

Reduction in salt and fat had a significant impact on the toughness of sausages which can be seen clearly in the left hand side of Figure 1. In Table 4, lower fat

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sausages are significantly (P>0.001) tougher than high fat sausages. Significant correlations between lower salt samples and toughness can be seen in Table 3; lower salt in sausages being significantly tougher has also been shown by Sheard, Hope, Hughes, Baker & Nute (2010) who ran a trial of commercial UK sausages, and found that sausages which contained higher salt levels were negatively correlated to skin toughness and firmness.

Juiciness can be described as the amount of moisture or juice that is perceived during mastication of a food (Hayes, 2009). Fats are known to help develop texture, mouthfeel and assist in the overall sensation of lubricity in foods (Giese, 1996). This explains the reason why juiciness was found to be more correlated to higher fat levels and higher salt as seen in the right side of Figure 1. Figure 2 also portrays correlations for juiciness to higher fat and higher salt products in its bottom right quadrant. Jack knifing results in Table 4 show these positive correlations (P<0.05) to juiciness and fat levels of 32.5% and 37.5%.

In Table 5, 2.4% salt is positively correlated (P>0.001) to juiciness with lower salt levels of 1.2%, 1% and 0.8% also being significantly negatively correlated. These correlations occurring between juiciness and higher salt level are most likely being created by increased levels of the myofibrillar proteins which are solubilised by the salt. More salt means increased solubilisation which in turn increases the water holding capacity and the perceived juiciness by the consumer.

Salt's distinctive taste in foods is primarily brought about by the Na⁺ cation in combination with the Cl⁻ anion effect on receptor cells (Miller & Bartoshuk, 1991; Murphy, Cardello & Brand, 1981). Sensory derived salt perception was found to be significantly correlated to salt levels above 1.6% (P>0.001) and negatively correlated to levels below 1.6% (P>0.001) in Table 4, this is in agreement with previous studies

(Ventanas et al. 2010; Ruusunen, Simolin & Puolanne, 2001; Matulis, McKeith, Sutherland & Brewer, 1995). These results indicate that small increases in salt level produce a subsequent significantly positive consumer detection level. Thus, consumers can clearly quantify salt levels in sausages without any major difficulty.

Fat level effect on salt can be seen in the right hemisphere of Figure 1 and the bottom right quadrant in Figure 2. Higher fat levels of 32.5% and 37.5% are more correlated to salt perception than the lower fat levels. Jack knifing from Table 4 shows significant correlations ((P>0.01) and (P>0.05), respectively) for the higher fat levels. Higher fat content in meats has also been shown in other studies to increase sensory salt perception (Tobin et al. 2012a; Ruusunen et al. 2001; Tuorila, Cardello & Lesher, 1994). Previously published data by Ruusunen et al. (2001) and Ruusunen, Vainionpaa, Lyly, Lahteenmaki, Niemisto & Ahvenainen (2005) postulate that the perceived salt intensity of a product is not solely based on the level of salt present, instead the background composition such as the lean meat content, plays a key role in salt perception when being assessed.

Consumer sensory scores on meat flavour (Figure 1) were shown to be directionally correlated to lower fat containing samples compared to higher fat containing samples which had significant negative correlations to meat flavour. The higher rating for meat flavour is likely due to the higher level of protein and moisture contents in these samples (Table 3). Meat flavour (Table 5) had significant but negative correlations to salt for the 2.4% salt (P<0.05), 2.0% (P<0.01) and for the 1.6% salt (P<0.001) containing sausages. This is likely due to the high correlation (P < 0.001) of the higher salt levels (2.4% and 2.0%) to salt perception which could impair a taster's ability to pick up the meat flavour.

Off-flavour was described to the consumer prior to tasting as, rancid, cardboard or linseed oil-like flavour. No significant correlations were found between levels of fat, however, high salt levels had significant negative correlations (P<0.05) to off-flavour. The lower values for off-flavour in high salt samples maybe due to the higher salt perception which could mask off-flavour present within samples.

Overall acceptability of samples was found to have no significant correlations when it came to fat levels, however, directional correlations exist showing a preference for lower fat samples (Table 4), this indicates that the level of fat can be reduced without any marked decrease in consumer acceptability. The ability to reduce fat in meat products without detrimental effects on consumer acceptability has also been reported by Crehan, Hughes, Troy & Buckley, (2000); Hughes, Cofrades & Troy, (1997) and Hughes, Mullen & Troy, (1998). Figure 2 does contain significant values which convey a negative correlation for 37.5% fat and 2.4, 2.0 and 1.6% salt samples to the overall acceptability. Salt levels in Table 5 show significant positive correlations for acceptability to lower salt levels of 1.4 % (P<0.001) and 1.0% (P<0.01) along with significant negative correlations to 2.4% (P<0.05), 2.0% (P<0.001) and 1.6% (P<0.05) salt.

5.3.2 Physiochemical analysis

Compositional analysis of the cooked samples is shown in Table 3 from this table samples produced with higher fat levels have a marked decrease in protein compared to lower fat samples. Samples with lower levels of salt also tended to contain more protein than the higher salt samples. Fat levels in Table 4 are highly correlated (P<0.001) to 37.5% and 32.5% fat samples. Samples with salt levels above 1.6% displayed a positive correlation to fat (P<0.01), where as lower salt levels of

0.8% had a significant negative correlation (P<0.001) to fat. Moisture content seems to be the inverse of fat, in that the lower fat samples correlated to higher moisture (P<0.001) in Table 4 and the higher salt samples correlated to lower moisture content (P<0.001) in Table 5.

Cooking loss in the samples is primarily made up of fat, and by comparing fat levels from Table 2 and Table 3 it can be seen that the higher fat samples lose a greater percentage (roughly 30%) of fat compared to the lower fat samples (roughly 15%). The lower fat sausages also contain more protein than higher fat sausages (Table 3). Cooking loss is shown to be correlated to higher fat in the top right quadrant of Figure 1. Table 2 also shows positive (P<0.001) significant correlations for 37.5% and 32.5% fat levels to cooking loss. Carballo, Mota, Barreto & Jimenez Colmenero (1995) found that sausages with more protein and less fat also had less cooking loss when measuring the total expressible fluid. Work carried out by Banon, Diaz, Nieto, Castillo & Alvarez (2008) showed that an increase in the fat to lean ratio in comminuted pork products caused a significant increase in cooking loss compared to lower fat products.

Table 5 indicates a higher cooking loss present in samples with salt levels of 1.6% and up with a significant (P<0.001) positive correlation. This correlation can also be seen in the right hemisphere of Figure 1. Figure 2 further portrays the effects of fat and salt on cooking loss showing that the higher fat levels with both high and low salt levels in the right-hand hemisphere are correlated to cooking loss, whereas low fat sausages with high salt are actually negatively correlated to cooking loss. These results indicate that the level of fat in samples' effect on cooking loss is far greater than the level of salts.

Colour in sausages is generally influenced by fat content and moisture (Ahmed, Miller, Lyon, Vaughters & Reagan, 1990) and also by the myoglobin and its state within the meat used to produce the product (Hand, Hollingswort, Calkins & Mandigo, 1987). Hunter Lab values of raw products showed that the L value was significantly higher (P<0.001) in higher in the 32.5% and 37.5% fat samples. Lower fat samples were shown to be darker in colour, this is in agreement with work done by Hand et al, (1987) who also found that lower fat samples had a darker colour than the higher fat samples.

Texture profile analysis of the samples as measured by a texture analyser has shown a range of correlations throughout the products. In Figure 1, hardness which is calculated as the peak force of the first compression had high positive correlations for low fat samples and lower salt samples. The anova values can be seen in Table 4 for fat with a (P<0.001) significance to 27.5% and 22.5% fat, and also in Table 5 for salt levels which shows a (P < 0.001) significance value to 1.2% and 1.0% salt levels. Lower fat levels in sausages have been documented as being tougher than higher fat sausages (Cengiz & Gokoglu, 2007; Ahmed et al. 1990; Hand et al. 1987). The decrease in fat coincidently causes an increase in protein content as seen in Table 3 which also tends to increase the moisture content of each product. This increase in moisture and protein, more specifically myofibrillar protein can create a denser and stable matrix within the product which then increases the hardness of the product (Colmenero, 1996; Cavestanty, Colmenero, Solas & Carballo, 1994)

The higher protein level in each product also affects other textural properties such as the springiness which can be explained as the products ability to return to its original dimensions after a deformation. Other studies have also shown an increase in springiness as fat was reduced (Barbut & Mittal, 1996; Gregg, Claus, Hackney & Marriott, 1993; Barbut & Mittal, 1992).

Cohesiveness and chewiness which are defined as how well the product withstands a second deformation relative to how it behaved under the first and the difficulty in chewing a product respectively. Low fat and low salt samples can be seen to correlate to these parameters in the upper left quadrant of Figure 1. These results are in agreement with the work done by Olivares, Navarro, Salvador & Flores (2010) who showed an increased level of cohesiveness and chewiness in lower fat ripened sausages than in higher fat sausages. Other authors who have reported the same results for increased chewiness in low fat products include García, Dominguez, Galvez, Casas & Selgas (2002) and Salazar, García & Selgas (2009).

5.4 Conclusions

Reduction in the levels of salt and fat in sausages had a range of effects on the final product. Lowering fat produced products which were rated to be less dark in colour, tougher, less juicy and taste less salty than higher fat products. Reduction in salt produced products which consumers found to be paler in colour, with increased tenderness and greater meat flavour than higher salt containing products.

In conclusion fat levels can be reduced in sausages without significantly reducing product quality and overall acceptability. Salt perception is important to consumer acceptability for sausages; lowering the level of salt too low (< 1.0 %) produces products that are unacceptable to the average consumer however producing a product within the limits of salt levels recommended by the FSAI is not only achievable but also can produce a superior product when rated by consumers.

5.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

5.6 Tables and Figures Table 1. Sausage Formulations:

	Composition					
Sample	% Fat	% Pork	% Salt	% Water	% Rusk	% Seasoning
1	37.50	26.60	2.40	20.00	12.50	1.00
2	37.50	27.00	2.00	20.00	12.50	1.00
3	37.50	27.40	1.60	20.00	12.50	1.00
4	37.50	27.60	1.40	20.00	12.50	1.00
5	37.50	27.80	1.20	20.00	12.50	1.00
6	37.50	28.00	1.00	20.00	12.50	1.00
7	37.50	28.20	0.80	20.00	12.50	1.00
8	32.50	31.60	2.40	20.00	12.50	1.00
9	32.50	32.00	2.00	20.00	12.50	1.00
10	32.50	32.40	1.60	20.00	12.50	1.00
11	32.50	32.60	1.40	20.00	12.50	1.00
12	32.50	32.80	1.20	20.00	12.50	1.00
13	32.50	33.00	1.00	20.00	12.50	1.00
14	32.50	33.20	0.80	20.00	12.50	1.00
15	27.50	36.60	2.40	20.00	12.50	1.00
16	27.50	37.00	2.00	20.00	12.50	1.00
17	27.50	37.40	1.60	20.00	12.50	1.00
18	27.50	37.60	1.40	20.00	12.50	1.00
19	27.50	37.80	1.20	20.00	12.50	1.00
20	27.50	38.00	1.00	20.00	12.50	1.00
21	27.50	38.20	0.80	20.00	12.50	1.00
22	22.50	41.60	2.40	20.00	12.50	1.00
23	22.50	42.00	2.00	20.00	12.50	1.00
24	22.50	42.40	1.60	20.00	12.50	1.00
25	22.50	42.60	1.40	20.00	12.50	1.00
26	22.50	42.80	1.20	20.00	12.50	1.00
27	22.50	43.00	1.00	20.00	12.50	1.00
28	22.50	43.20	0.80	20.00	12.50	1.00

Raw Composition										
Sample	% Moist	ure	% Fat		% Protein		% Ash		% Carbohydrate	
F 37.5% S 2.4%	41.65 ±	0.17	$36.87~\pm$	0.06	6.12 ±	0.103	$2.88~\pm$	0.03	12.48 \pm	0.002
F 37.5% S 2.0%	$41.41~\pm$	0.14	$37.24~\pm$	0.04	$6.21~\pm$	0.103	$\textbf{2.48}~\pm$	0.02	$12.66~\pm$	0.001
F 37.5% S 1.6%	$41.43~\pm$	0.11	$37.44~\pm$	0.01	$6.30~\pm$	0.101	$2.09~\pm$	0.02	12.74 \pm	0.001
F 37.5% S 1.4%	$41.43~\pm$	0.17	$37.98~\pm$	0.02	$6.35~\pm$	0.069	$1.88~\pm$	0.05	$12.36~\pm$	0.002
F 37.5% S 1.2%	$42.08~\pm$	0.28	$37.59~\pm$	0.02	$6.39~\pm$	0.155	$1.69~\pm$	0.03	$12.25~\pm$	0.001
F 37.5% S 1.0%	$42.36~\pm$	0.23	$37.22~\pm$	0.09	$6.44~\pm$	0.117	$1.50\ \pm$	0.02	$12.48~\pm$	0.002
F 37.5% S 0.8%	$41.62~\pm$	0.39	$37.61~\pm$	0.05	$6.49~\pm$	0.101	$1.30~\pm$	0.03	$12.98~\pm$	0.001
F 32.5% S 2.4%	$44.80~\pm$	0.21	$32.55~\pm$	0.02	$7.27~\pm$	0.112	$2.93~\pm$	0.03	$12.45~\pm$	0.003
F 32.5% S 2.0%	$44.87~\pm$	0.08	$32.68~\pm$	0.06	$7.36~\pm$	0.155	$2.54~\pm$	0.04	$12.55~\pm$	0.001
F 32.5% S 1.6%	$45.69~\pm$	0.19	$31.98~\pm$	0.03	$7.45~\pm$	0.175	$2.14~\pm$	0.04	12.74 \pm	0.001
F 32.5% S 1.4%	$45.83~\pm$	0.43	$32.35~\pm$	0.02	$7.50\ \pm$	0.084	$1.96~\pm$	0.04	$12.36~\pm$	0.001
F 32.5% S 1.2%	$44.98~\pm$	0.44	$32.88~\pm$	0.01	$7.54~\pm$	0.14	$1.75 \pm$	0.03	$12.85~\pm$	0.002
F 32.5% S 1.0%	$45.55~\pm$	0.30	$32.56~\pm$	0.06	$7.59~\pm$	0.112	$1.55 \pm$	0.04	12.75 \pm	0.001
F 32.5% S 0.8%	$45.76~\pm$	0.20	$32.32~\pm$	0.03	$7.64~\pm$	0.138	1.33 \pm	0.15	$12.95~\pm$	0.001
F 27.5% S 2.4%	$48.45~\pm$	0.21	$27.69~\pm$	0.20	$8.42~\pm$	0.071	$2.99~\pm$	0.02	$12.45~\pm$	0.001
F 27.5% S 2.0%	$48.84~\pm$	0.19	$27.45~\pm$	0.05	$8.51\ \pm$	0.079	$2.54~\pm$	0.07	$12.66~\pm$	0.001
F 27.5% S 1.6%	$48.76~\pm$	0.17	$28.00~\pm$	0.03	$8.60\ \pm$	0.288	$2.20~\pm$	0.02	12.44 \pm	0.002
F 27.5% S 1.4%	$49.97~\pm$	0.18	$27.26~\pm$	0.05	$8.65~\pm$	0.163	$2.00~\pm$	0.05	12.12 \pm	0.002
F 27.5% S 1.2%	$49.33~\pm$	0.18	$27.88~\pm$	0.02	$8.69~\pm$	0.113	$1.80~\pm$	0.03	$12.30~\pm$	0.001
F 27.5% S 1.0%	$49.67~\pm$	0.15	$27.41~\pm$	0.08	$8.74~\pm$	0.296	1.61 \pm	0.02	$12.57~\pm$	0.001
F 27.5% S 0.8%	$49.31~\pm$	0.12	$27.61\ \pm$	0.06	$8.79~\pm$	0.114	$1.39~\pm$	0.03	$12.90~\pm$	0.001
F 22.5% S 2.4%	$52.68~\pm$	0.25	$22.13~\pm$	0.02	$9.57~\pm$	0.06	$3.04~\pm$	0.04	$12.58~\pm$	0.001
F 22.5% S 2.0%	$52.77~\pm$	0.15	$22.44~\pm$	0.04	$9.66~\pm$	0.071	$2.66~\pm$	0.03	$12.47~\pm$	0.004
F 22.5% S 1.6%	$52.79~\pm$	0.16	$22.58~\pm$	0.02	$9.75~\pm$	0.249	$2.25~\pm$	0.05	12.63 \pm	0.002
F 22.5% S 1.4%	$52.76~\pm$	0.16	$22.77~\pm$	0.08	$9.80~\pm$	0.148	$2.05~\pm$	0.05	$12.62~\pm$	0.002
F 22.5% S 1.2%	$53.06~\pm$	0.12	$22.86~\pm$	0.03	$9.84~\pm$	0.144	$1.88~\pm$	0.02	$12.36~\pm$	0.002
F 22.5% S 1.0%	$53.03~\pm$	0.18	$22.54~\pm$	0.04	$9.89~\pm$	0.299	$1.66~\pm$	0.02	$12.88~\pm$	0.002
F 22.5% S 0.8%	$53.85~\pm$	0.16	22.27 ±	0.04	$9.94~\pm$	0.079	1.44 \pm	0.08	$12.50~\pm$	0.001
E - Eat										

Table 2. Raw sausage composition.

F - Fat S - Salt

Table 3. Cooked sausage composition.

	Cooked Composition									
Sample	% Moist	% Moisture % Fat % Pr		% Prote	ein	in % Ash		% Carbohydrate		
F 37.5% S 2.4%	$49.40~\pm$	0.08	$28.18~\pm$	0.08	10.32 \pm	0.04	$3.25 \pm$	0.03	$8.85 \pm$	0.08
F 37.5% S 2.0%	$49.86~\pm$	0.06	$27.18~\pm$	0.04	10.43 \pm	0.05	$2.85~\pm$	0.04	$9.68 \pm$	0.03
F 37.5% S 1.6%	$47.97~\pm$	0.01	$27.95~\pm$	0.02	10.43 \pm	0.09	$2.46~\pm$	0.04	11.19 \pm	0.03
F 37.5% S 1.4%	$48.28~\pm$	0.02	$27.34~\pm$	0.03	10.53 \pm	0.02	$2.25~\pm$	0.08	11.61 \pm	0.03
F 37.5% S 1.2%	$48.22~\pm$	0.06	$27.25~\pm$	0.09	$10.81~\pm$	0.08	$2.06~\pm$	0.03	$11.66~\pm$	0.04
F 37.5% S 1.0%	$47.32~\pm$	0.04	$27.59~\pm$	0.05	$10.58~\pm$	0.07	$1.87~\pm$	0.03	$12.64~\pm$	0.04
F 37.5% S 0.8%	$48.30\ \pm$	0.02	$27.84~\pm$	0.01	$10.81~\pm$	0.05	$1.67~\pm$	0.03	$11.38~\pm$	0.05
F 32.5% S 2.4%	$50.12~\pm$	0.03	$26.56\ \pm$	0.07	11.45 \pm	0.08	$3.30~\pm$	0.08	$8.57~\pm$	0.07
F 32.5% S 2.0%	$50.39~\pm$	0.06	$26.09~\pm$	0.05	$11.67~\pm$	0.05	$2.91~\pm$	0.08	$8.94~\pm$	0.06
F 32.5% S 1.6%	$48.03~\pm$	0.03	$26.34~\pm$	0.05	$11.99~\pm$	0.09	$2.51~\pm$	0.05	$11.13~\pm$	0.08
F 32.5% S 1.4%	$48.23~\pm$	0.02	$26.65\ \pm$	0.00	$11.87~\pm$	0.08	$2.33~\pm$	0.04	$10.92~\pm$	0.02
F 32.5% S 1.2%	$47.92~\pm$	0.01	$26.64\ \pm$	0.01	11.76 \pm	0.01	$2.12~\pm$	0.03	$11.56~\pm$	0.07
F 32.5% S 1.0%	$48.95~\pm$	0.04	$25.41\ \pm$	0.03	11.98 \pm	0.07	$1.92~\pm$	0.06	11.74 \pm	0.04
F 32.5% S 0.8%	$49.38~\pm$	0.02	$25.14\ \pm$	0.07	$12.03~\pm$	0.08	$1.70~\pm$	0.04	11.75 \pm	0.04
F 27.5% S 2.4%	$53.45~\pm$	0.09	$22.01\ \pm$	0.05	$12.97~\pm$	0.08	$3.36~\pm$	0.07	$8.21~\pm$	0.03
F 27.5% S 2.0%	$52.89~\pm$	0.04	$21.82~\pm$	0.07	$13.05~\pm$	0.04	$2.91~\pm$	0.04	$9.33~\pm$	0.08
F 27.5% S 1.6%	$50.36~\pm$	0.05	$22.99~\pm$	0.04	$13.09~\pm$	0.07	$2.57~\pm$	0.04	$11.00~\pm$	0.08
F 27.5% S 1.4%	$51.62~\pm$	0.03	$22.53~\pm$	0.03	13.23 \pm	0.04	$2.37~\pm$	0.08	$10.25~\pm$	0.05
F 27.5% S 1.2%	52.17 \pm	0.08	$22.06~\pm$	0.06	13.23 \pm	0.09	$\textbf{2.17}~\pm$	0.03	$10.37~\pm$	0.04
F 27.5% S 1.0%	52.33 \pm	0.05	$22.04~\pm$	0.06	13.81 \pm	0.06	$1.98~\pm$	0.03	$9.84~\pm$	0.03
F 27.5% S 0.8%	52.33 \pm	0.03	$22.39~\pm$	0.04	13.65 \pm	0.02	$1.76~\pm$	0.03	$9.87~\pm$	0.03
F 22.5% S 2.4%	$55.86~\pm$	0.20	$19.56~\pm$	0.07	15.01 \pm	0.07	$3.41~\pm$	0.05	$6.16~\pm$	0.03
F 22.5% S 2.0%	$55.76~\pm$	0.06	$18.60~\pm$	0.11	15.17 \pm	0.07	$3.03~\pm$	0.07	$7.44~\pm$	0.07
F 22.5% S 1.6%	$55.00\ \pm$	0.02	$19.03~\pm$	0.01	$15.28~\pm$	0.09	$2.62~\pm$	0.05	$8.06~\pm$	0.05
F 22.5% S 1.4%	54.73 \pm	0.05	19.13 \pm	0.02	15.33 \pm	0.10	$\textbf{2.42}~\pm$	0.06	$8.39~\pm$	0.08
F 22.5% S 1.2%	$54.38~\pm$	0.02	19.41 \pm	0.01	15.04 \pm	0.02	$2.25~\pm$	0.08	$8.92~\pm$	0.03
F 22.5% S 1.0%	$54.92~\pm$	0.04	$19.37~\pm$	0.04	$15.68~\pm$	0.01	$2.03~\pm$	0.02	$8.00\ \pm$	0.06
F 22.5% S 0.8%	$56.27~\pm$	0.02	$18.97~\pm$	0.02	15.55 ±	0.04	1.81 ±	0.07	$7.40~\pm$	0.04
E Ect										

F - Fat S - Salt

Attribute	22.5% Fat	27.5% Fat	32.5% Fat	37.5% Fat
Colour ^a	-0.1637ns	-0.0256*	0.0001***	0.0002***
Texture ^a	0.5309ns	0.9114ns	-0.0333*	-0.0046**
Toughness ^a	0.0001***	0.9001ns	-0.6283ns	-0.0001***
Juiciness ^a	-0.0001***	-0.8375ns	0.0453*	0.0156*
Salt Taste ^a	-0.0092**	-0.0252*	0.0061**	0.0226*
Meat Flavour ^a	0.7623ns	0.0795ns	-0.0369*	-0.0012**
Off Flavour ^a	0.0075ns	0.1384ns	-0.6281ns	-0.0001***
Acceptability ^a	0.7481ns	0.0525ns	-0.2068ns	-0.9908ns
Cooking Loss ^b	-0.0001***	-0.0001***	0.0001***	0.0001***
Cooked L Value ^b	0.0001***	0.0001***	-0.0001***	-0.0001***
Cooked a Value ^b	-0.0001***	-0.0001***	0.0001***	0.0001***
Cooked b Value ^b	-0.0069**	-0.0004***	0.0254*	0.0001***
Raw L Value ^b	-0.9153ns	-0.1453ns	0.0001***	0.0001***
Raw a Value ^b	0.0001***	0.0533ns	-0.0001***	-0.0011**
Raw b Value ^b	0.1214ns	0.7883ns	-0.0001***	-0.0001***
Moisture Content	t 0.0001***	0.0001***	-0.0001***	-0.0001***
Fat Content ^b	-0.0001***	-0.0001***	0.0001***	0.0001***
Hardness ^b	0.0001***	0.0001***	-0.0001***	-0.0001***
Springiness ^b	0.0016**	0.0001***	-0.0001***	-0.0001***
Cohesiveness ^b	0.0001***	0.0001***	-0.0001***	-0.0001***
Chewiness ^b	0.0001***	0.0001***	-0.0001***	-0.0001***
Resilience ^b	0.0001***	0.0001***	-0.0001***	-0.0161**

Table 4. P-values of estimated regression coefficients (ANOVA values) for the relationships of sensory terms ^a and instrumental ^b measurements as derived by Jack-knife uncertainty testing for sausages with varying fat levels.

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c P values of estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates weather the correlation is positively or negatively correlated. ^d Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01;

^a Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01; ***=P<0.001.
Table 5. P values of estimated regression coefficients (ANOVA values) for the relationships of sensory terms ^a and instrumental ^b measurements as derived by Jack-knife uncertainty testing for sausages with varying salt levels.

Attribute	2.4 % Salt	2.0% Salt	1.6% Salt	1.4% Salt	1.2% Salt	1.0% Salt	0.8% Salt
Colour ^a	0.1583ns	0.2978ns	0.4302ns	-0.1062ns	-0.0008***	-0.3419ns	-0.0458*
Texture ^a	-0.0108*	-0.0007***	-0.8887ns	0.0151*	0.0001***	0.0736ns	0.1541ns
Toughness ^a	-0.9557ns	-0.0127*	-0.4429ns	0.0188*	0.0054**	0.9383ns	0.0001***
Juiciness ^a	0.0006***	0.0711ns	0.3207ns	-0.7086ns	-0.0396*	-0.0335*	-0.0001***
Salt Taste ^a	0.0001***	0.0001***	0.1647ns	-0.0001***	-0.0001***	-0.0001***	-0.0001***
Meat Flavour ^a	-0.0273*	-0.0056**	-0.0001***	-0.0099ns	0.0864ns	0.0156*	0.4499ns
Off Flavour ^a	-0.0351*	-0.1661ns	-0.0191*	0.2265ns	0.1161ns	0.0172*	0.6021ns
Acceptability ^a	-0.0129*	-0.0001***	-0.0132*	0.0002***	0.0631ns	0.0032**	0.8886ns
Cooking Loss ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0008***	-0.0001***	-0.0001***
Cooked L Value ^b	-0.5861ns	-0.0054**	-0.0711ns	0.2229ns	0.9449ns	0.7781ns	0.2107ns
Cooked a Value ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***	-0.0001***	-0.0001***
Cooked b Value ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0787ns	-0.0001***	-0.0001***
Raw L Value ^b	0.0261*	0.0001***	0.0001***	-0.5376ns	-0.0001***	-0.6819ns	-0.0014**
Raw a Value ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***	0.0001***	0.0001***	0.0001***
Raw b Value ^b	-0.0018**	-0.0001***	-0.035*	0.0111*	0.0001***	0.3674ns	0.1005ns
Moisture Content ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***	0.0001***	0.0003***	0.0949ns
Fat Content ^b	0.0001***	0.0003***	0.0069**	-0.2625ns	-0.1469ns	-0.0517ns	-0.0001***
Hardness ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***	0.0001***	0.0001***	0.0539ns
Springiness ^b	-0.5356ns	-0.0001***	-0.4443ns	0.0001***	0.0001***	0.8954ns	0.0007***
Cohesiveness ^b	-0.0001***	-0.0001***	-0.2077ns	0.0001***	0.0001***	0.0004***	0.0311*
Chewiness ^b	-0.0001***	-0.0001***	-0.0001***	0.0001***	0.0001***	0.0001***	0.0806ns
Resilience ^b	-0.0001***	-0.0024**	-0.3189ns	0.2282ns	0.0796ns	0.0439*	0.0001***

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c P values of estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates weather the correlation is positively or negatively correlated. ^d Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01;

^a Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01; ***=P<0.001.

Figure 1. ANOVA-partial least squares regression (APLSR) correlation loading plot for each of the fat and salt sausage treatment groups.



Shown are the loadings of the X- and Y-variables for the first 2 PCs for \blacktriangle = the salt % and fat %, = sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 35.77% and the calibrated variance was 36.37%.





Shown are the loadings of the X- and Y-variables for the first 2 PCs for=the individual treatments, \bullet =sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 32.73% and the calibrated variance was 33.40%.

Chapter 6: Effect of Varying Salt and Fat Levels on the Sensory and Physiochemical Quality of Frankfurters

This Chapter is in the form of a manuscript accepted for publication in *Meat Science* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Effect of varying salt and fat levels on the sensory and physiochemical quality of frankfurters. *Meat Science*, 92, 659-666.

Abstract

The sensory and physiochemical properties of Frankfurters with varying fat and salt levels were investigated. Twenty Frankfurters formulations were produced with varying concentrations of fat (10%, 15%, 20%, 25% w/w) and salt (1%, 1.5% 2%, 2.5%, 3% w/w). Frankfurters were assessed instrumentally for colour, moisture, fat, cooking loss and texture profile analysis. Consumers (n=25) evaluated each product for colour, coarseness, tenderness, juiciness, salt taste, meat flavour, off-flavour and overall acceptability using a hedonic scale. Salt levels below 1.5% were shown to have a negative effect on consumer acceptability, with 2.5% salt concentration being the most accepted (P<0.001) by consumers. However, Frankfurters containing the lower fat levels 10% and 15% fat with higher salt levels (2.5-3%) were significantly the most acceptable variants to consumers. Samples containing less fat and salt were found to be tougher, less juicy and had greater cooking losses. Thus salt perception is very important for consumer acceptability, but fat levels can be potentially reduced without significantly affecting overall acceptability.

6.1 Introduction

In recent years consumer studies have begun to show that meat consumption is being more and more influenced by health and nutritional considerations (Fonseca & Salay, 2008; Angulo & Gil, 2007). Hypertension due to high blood pressure is a growing concern for many countries worldwide as it has been linked to cardiovascular disease CVD (Morgan et al. 2001; Sacks et al. 2001; Tuomilehto et al. 2001). In China alone hypertension has tripled since 1958, and CVD claims as many as 2.6 million lives every year (Xiaosong, 2007). The estimated cost of cardio vascular disease to the EU economy has been estimated at €169 billion every year (Petersen et al., 2005), similar figures are shown in the USA where CVD related illness has an estimated cost of \$403.1 Billion/year (Thom et al., 2006).

Links between an excessive intake of sodium to hypertension has been shown in many studies (Law, Frost, & Wald, 1991a, 1991b; Dahl, 1972). Salt (NaCl) is the main source of sodium in our diets, 75% of our dietary intake comes from processed foods (Appel & Anderson, 2010). The total dietary salt intake has been recommended at 5-6 g/day (Aho et al. 1980; WHO, 1990; Desmond, 2006). In many industrialised European countries this recommended dietary intake is greatly exceeded and has been estimated to be as high as 9–12 g NaCl/day (Intersalt Cooperative Research Group, 1988).

High fat levels in our diets also have been shown to have adverse health effects promoting CVD, obesity, diabetes and certain types of bowel cancer (Aggett et al. 2005). Obesity levels amongst the general population have shown to be greater than 30% in the United States and between 15 and 20% in Britain and Ireland (Bassett et al. 2008). In 2000 it was called "the greatest health threat facing the West" by the World Health Organisation. The recommended dietary intake of fat should be no more

than 20 to 35% of ones total calorific intake (National Academy of Sciences. Institute of Medicine. Food and Nutrition Board, 2011). Fats, however, play an important role in meat products. They interact with other ingredients and help to develop texture, mouthfeel and provide a lubricating effect in processed meats, as well as contributing to overall flavour (Javidipour et al. 2005; Crehan et al. 2000; Giese, 1996).

Salt works as a preservative in food because of its osmotic effect in high concentrations and its ability to lower the water activity value in food which in turn slows down or even stops vital microbial processes. The perceived taste of salt in foods is primarily due to the Na⁺ cation in combination with Cl⁻ anion effect on receptor cells (Miller & Bartoshuk, 1991; Murphy et al. 1981). Water holding capacity is the ability of a food to trap water within a three dimensional structure (Chantrapornchai & McClements, 2002). In meat products the salt is used to extract myofibrillar proteins which associate into a gel when heated (Foegeding & Lanier, 1987). This gel network in turn increases the water holding capacity of the meat product.

Comminuted (ground) cooked meat products (gel/emulsion systems) are a commercially important group of meat products, of which Frankfurters are among one of the more popular varieties. Frankfurters are frequently consumed meat products which are of considerable economic importance and enjoy wide consumer acceptance in certain sections of the global population (Delgado-Pando et al. 2010). Frankfurters are a type of highly seasoned sausage, traditionally comprised of mixed pork and beef. Frankfurters can contain up to 30% fat with an industrial average of about 20% (Keeton, 1994) and high salt concentrations ranging from 2% and higher. Research on available products is essential to develop healthier, lower-cost alternatives to Frankfurters currently marketed (González-Viňas, Caballero, Gallego & García Ruiz,

2004). Several studies have been conducted which have investigated ways in which healthier Frankfurters can be manufactured (Delgado-Pando et al. 2010, 2011; Jiménez-Colmenero et al. 2010; López-López, Cofrades & Jiménez-Colmenero, 2009; Ayo et al. 2007; González-Viňas, 2004; Paneras & Bloukas, 1994; Bloukas & Paneras, 1993; Márquez, Ahmed, West & Johnson, 1989; Park, Rhee, Keeton & Rhee, 1989), but none of these studies have explored the matrix interaction of salt and fat and how these parameters can be manipulated to produce a healthier consumer optimised Frankfurter product. Additionally, there is increasing demand by consumers for 'clean label' products without the requirement for use of additives and replacers. Consumers associate reduced fat foods with inferior sensory properties and perceive them with a degree of scepticism and mistrust and that there is a concern about substitutes and additives used to replace fat. This implies that there is more to reduced fat product acceptance than sensory experience (Hamilton, Knox, Hill & Parr, 2000). There is evidence to suggest that consumers are also sceptical of health and nutrition claims on packages viewing the claims as attempts by the manufacturer to sell more of their product (Levy & Stokes, 1987). Nonetheless, it is imperative to attempt to establish the limit at which salt and fat can be eliminated or reduced from processed meat products to enhance health status, but not in a manner that would negatively impact on functionality, product quality or adversely affect sensory or consumer acceptability.

The objective of this study was to investigate interactions between different salt and fat levels on the overall quality of cooked Frankfurters and also to investigate the consumer optimisation of reduced salt and fat variants, without using fat and salt alternatives.

6.2 Materials and Methods

6.2.1 Sample Preparation

Beef and pork were selected on the basis of a high visual lean (V/L) score, (FSA, 2003); pork shoulder was used with a V/L score of 99% and beef shin was used with a V/L score of 95%. Beef and pork were purchased along with pork back fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The meat and fat were vacuum packed and stored at -18°C until required for Frankfurter production. The frozen meat and fat was then cut into strips and allowed to thaw slightly before being minced through a 3mm plate (TALSABELL S. A., Spain). The meat was weighed according to the formulations shown in Table 1 and fed into the bowl chopper. The required salt, nitrite and two thirds of the water were added and mixed at high speed (3000 rpm.) for 60 seconds. The required fat and seasoning was then added and the mix was chopped for further 120 seconds at high speed. The remaining water was then added and mixed at high speed for 45 seconds. The Frankfurter mix was then placed into the casing filler and stuffed into cellulose casings. The Frankfurters were hung in a Zanussi convection oven (C. Batassi, Conegliano, Italy) and cooked at 90°C until an internal temperature of 72°C tested by a temperature probe (Testo 110, Lenzkirch, Germany) was reached, the Frankfurters were then held at 72°C for 10 minutes. All test samples were cooked at the same time and segregated to prevent any mixing. The Frankfurters were then sealed into polyamide/polyethylene (PA/PE) laminate plastic bags and stored in the chill over night at -4°C.

6.2.2 Sensory Evaluation

Sensory analysis using 25 consumers in the age range 20–30 years was performed, following the method by O'Sullivan, Byrne & Martens (2003). All were selected on the basis that they consume and purchase Frankfurter meat products regularly. For each Frankfurter, consumers were asked to indicate their score on a 10 cm line scale ranging from 0 at the left to 10 at the right and rating subsequently scored in cm from left. The consumers were asked to evaluate the Frankfurters using the following descriptors: colour, coarseness, toughness, juiciness, springiness, salt taste, meat flavour, off-flavour and overall acceptability. Off-flavour was described to consumers as off-flavour, rancid, cardboard or linseed oil-like flavour. Sensory analysis was undertaken in the panel booths at the university sensory laboratory that conforms to ISO (1988) international standard. Five samples were presented to the consumers and they were required to rinse with water before tasting each sample. Sample presentation order was randomised to prevent any flavour carryover effects (MacFie, Bratchell, Greenhoff & Vallis, 1989). All analysis was undertaken in duplicate.

6.2.3 Protein Content

The Kjeldahl method (Suhre, Corrao, Glover & Malanoski, 1982) was used to measure protein concentrations. The digestion block was pre-heated to 410 °C. Approximately 0.5 g of well homogenised sample was weighed accurately into a digestion tube. 15 ml of sulphuric acid (nitrogen free), 10ml hydrogen peroxide and 2 "kjeltabs" were added to the sample. The tubes where then inserted in the heated digestion block. When the samples became colourless they were removed from the block. The tubes were allowed to cool in the fume hood after removal.

50 ml of distilled water was carefully added to the cooled and digested sample inside the fume-hood. The tubes and a receiver flask containing 50 ml of 4 % Boric acid with indicator were then placed into the distillation unit. After the sample had been distilled the contents of the receiver flask was titrated against 0.1 N hydrochloric acid until the green colour reverted back to the original red colour.

6.2.4 Ash Content

Ash content was determined using a muffle furnace (AOAC, 1923). A muffle furnace was pre-heated to 525°C. Approximately 5 g of well homogenised sample was weighed into porcelain dishes using a balance that weighs to 1 mg. The dishes containing samples were then put in the muffle furnace for approximately 6 hours until the colour of the samples went white. The dishes containing the samples were then removed and placed in a desiccator to cool. The dishes were then weighed and the ash content calculated.

6.2.5 Moisture and Fat Content

A total of 200g of Frankfurter sample was homogenised using a Büchi Mixer B-400 (BÜCHI Labortechnik AG, Meierseggstrasse 40, Postfach, CH-9230 Flawil 1, Switzerland) and quickly transferred into a moisture proof bag to avoid moisture or evaporative loss. Moisture and fat content were then determined using the CEM SMART (moisture) and SMART Trac (fat) systems (Bostian et al. 1985). All analysis was undertaken in triplicate.

6.2.6 Colour

The cooked Frankfurters were cut down the centre and were measured according to the CIE L* a* b* colour system using a Minolta CR 300 colorimeter (Minolta Camera Co. Ltd., Osaka, Japan) with an 11 mm-diameter aperture, D_{65} illuminant, calibrated by the CIE Lab colour space system using a white tile (C: Y = 93.6, x = 0.3130, y = 0.3193), (Minolta calibration plate). The cooked Frankfurters were split down the middle and colour measurements (CIE L*, a* and b* values representing lightness, redness and yellowness, respectively) were taken after samples were cooled to room temperature. Nine readings were taken per sample.

6.2.7. Cooking Loss

A total of nine Frankfurters sample from each formulation had their weights recorded before and after cooking and the differences in weights were recorded. The Frankfurters were hung in a Zanussi convection oven and cooked at 90°C until an internal temperature of 72°C tested by a temperature probe (Testo 110, Lenzkirch, Germany) was reached, the Frankfurters were then held at 72°C for 10 minutes. Before weighing, samples were blotted with a paper towel to remove excess surface moisture.

Calculation for cook loss was as follows:

% cook loss = (cooked weight – raw weight)/raw weight)) x 100

6.2.8 Texture Analysis

Texture measurements in the form of texture profile analysis (TPA) were performed at room temperature with a Texture Analyser 16 TA-XT2i (Stable Micro Systems, Surrey, UK). Three (10 mm x 10 mm) cylindrical patties were taken and subjected to a two-cycle compression test using the 25kg load cell. The samples were compressed to 40% of their original height with a cylindrical probe (SMSP/35 Compression plate) 35 mm diameter and a cross-head speed of 1.5 mm/s. Texture profile parameters were determined following descriptions by Bourne (1978) and the SMS manual (Stable Micro Systems, Surrey, UK), these included; hardness (N) maximum force required to compress the sample, springiness (m), ability of the sample to recover its original form after deforming force was removed, adhesiveness ($N \times s$), area under the abscissa after the first compression; cohesiveness, extent to which the sample could be deformed prior to rupture; and chewiness (J), work required to masticate the sample before swallowing. All analyses were performed in triplicate.

6.2.9 Data Analysis

ANOVA-partial least squares regression (APLSR, Fig. 1) was used to process the raw data accumulated from the 25 test subjects during the sensory evaluation and data acquired by instrumental methods. The X-matrix was designed as 0/1 design variables for fat and salt content of Frankfurters. The Y-matrix was designed as sensory, chemical, and instrumental variables. The optimal number of components in the APLSR models presented was determined to be four principal components (Fig. 1). PC 1 versus PC 2 is presented; the other PCs did not yield additional information or provide any predictive improvement in the Y-matrix obtained through their examination. The validated explained variance for the model constructed was 42.93% and the calibrated variance was 42.21%. For Figure 2 PC1 versus PC2 is also presented with a validated explained variance of 47.43% and a calibrated variance of 48.91%. To derive significance indicators for the relationships determined in the quantitative APLSR, regression coefficients were analysed by Jack-knifing (Table 4) which is based on cross-validation and stability plots (Martens & Martens, 1999, 2001). A second APLSR plot was constructed, identical to Figure 1, except instead of plotting the main effects for Fat and Salt the individual product combinations of fat and salt (n=20) were plotted against the sensory evaluation and data acquired by instrumental methods in order to investigate any additional interactions not observed in the first APLSR. All analyses were performed using the Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway).

6.3 Results and Discussion

6.3.1 Sensory Consumer Evaluation

Sensory evaluation and instrumental data are presented in the APLSR plot in Figure 1 with the corresponding regression co-efficient in Table 4 for fat and Table 5 for salt effects. Colour, as determined by the 25 consumers, was measured from extremely dark to extremely pale and from Fig. 1 was found to be positively correlated to higher fat levels and negatively correlated to lower fat levels (10% and 15%), whereas higher fat samples (20% and 25%) were perceived by the consumers to be lighter in colour. Lower salt levels such as 1.5% and 1% were found to be more positively correlated to a darker/deeper colour when compared to the higher salt samples. These findings are similar to those reported by Ventanas et al. (2010) who showed that a more intense colour was perceived by consumers in sausages containing higher salt levels than those containing lower salt levels.

From Table 4 and Figure 1 it can be clearly observed that none of the fat level variants assessed were significantly correlated to sensory toughness, however, the 10% fat level displayed a directional positive correlation to sensory toughness. Thus, fat levels used in the present study did not deleteriously affect consumer perception of toughness.

The level of salt in samples can be seen to have had a significant effect on the coarseness and toughness of Frankfurters. Figure 1 and Table 5 show significant and positive correlations to sensory toughness (P<0.01) and coarseness (P<0.05) for 1.5% salt containing samples and a positive but non-significant directional correlation for the 1% salt samples. Thus, consumers found these samples to be tougher when compared to the higher salt containing samples such as 2% and 3% salt. Both these samples were negatively and significantly (P<0.01) correlated to sensory toughness. Samples containing 1% and 1.5% salt were also significantly (P<0.001) correlated to cooking loss (Table 5). Thus, the cooking losses resulted in increased cook out and consumer perceived toughness in these samples.

From Figure 1 juiciness is seen to be negatively correlated to 10% fat and more highly attributed to the higher fat containing samples. In Table 4 it can be seen that 10% fat Frankfurters are significantly (P<0.05) negatively correlated to juiciness, whereas 25% and 15% fat Frankfurters are significantly (P<0.01) and positively correlated to juiciness. Similar findings on the effect of fat levels on juiciness sensory scores for beef patties can be seen in work by Berry & Leddy (1984) and Tobin et al. (2012a) and by work carried out by Mittal & Barbut (1994) in low fat Frankfurters.

The level of salt in Frankfurters had a high impact on the sensory perception of juiciness. Figure 1 and Table 5 display the high positive correlations (P<0.001) between juiciness and higher salt levels (2.5 and 3.0%) in Frankfurters, whereas significant negative correlations (P<0.001) can be observed between juiciness and lower salt levels (1.5% and 1.0%) in frankfurters. Ruusunen et al. (2002) showed similar results where higher salt Frankfurters had increased firmness and juiciness. The correlations between juiciness and higher salt levels is most likely due to the greater extraction of myofibrillar proteins, thereby leading to an enhanced moisture binding effect in these Frankfurters.

Salt perception in Frankfurters was clearly shown to be highly correlated to the higher salt containing Frankfurters. In Figure 1 and for the regression co-efficient shown in Table 5 significant (P<0.001) positive correlations for 2.5% and 3.0% salt levels and (P<0.001) negative correlations for 1.0% and 1.5% salt levels were observed for sensory-derived salt perception. Salt concentration effects on salt perception showed that small increases in salt level displayed a subsequent significantly positive consumer detection level. Thus, consumers can clearly quantify salt levels in Frankfurters without any great difficulty.

The bottom quadrant in Figure 1 displays the effect of fat content on salt perception, showing that the samples with 10% fat are perceived to be less salty. Additionally the regression co-efficient shown in Table 4 demonstrate a negative (P< 0.001) correlation between 10% fat and salt perception. The other samples displayed positive correlations to salt taste, the 15% and 20% samples significantly (P< 0.05) so. In the present study the 10% fat Frankfurters have an increased aqueous phase compared to other variants, thus diluting salt perception as the salt is dissolved in this phase. Similar effects linking lower fat content samples with lower salt perception

scores have been shown previously by Matulis et al. (1995) and Ruusunen et al. (2005) who both postulated that the effect of meat content on perceived saltiness in Frankfurters, as well as beef and pork meat patties, had a stronger effect on perceived saltiness than the level of fat present in the product. The results from the present study are also in agreement with that presented by Pappa, Bloukas & Arvanitoyannis (2000), who showed that the higher the salt level used in product formulation, the higher the perceived saltiness in the low-fat Frankfurters. Thus, the reduction in fat content decreases perceived salt taste.

Meat flavour scores, (Figure 1), were shown to be directionally correlated to high fat containing samples compared to lower fat containing samples. From Table 1, meat flavour was negatively and significantly (P<0.05) correlated to 10% fat Frankfurters and positively and significantly (P<0.001) correlated to 20% fat Frankfurters. Fat is also known to be a flavour enhancer. Fat interacts with other ingredients and helps to develop texture, mouthfeel and provide a lubricating effect in processed meats as well as contribute to the overall flavour (Javidipour et al. 2005; Crehan et al. 2000; Giese, 1996). This explains the positive correlation between increased fat content to perceived meat flavour by consumers.

Meat flavour (Table 5) can be seen to be significantly and positively correlated to salt for the 2.5% salt (P<0.001) and for the 3.0% salt (P<0.05) containing Frankfurters. On the other hand a negative correlation (P<0.001) is observed for the 1.5% salt containing Frankfurters. The salt acts as a flavour enhancer for natural meat flavour present in the samples. Salt is used in the production of many meat products for its role as a preservative, flavour enhancer (Silva et al. 2003) and its influence on water holding capacity (Lawrence et al. 2003).

Off-flavour was described to consumer prior to tasting as, rancid, cardboard or linseed oil-like flavour. Off-flavour (oxidative) scores as presented in Figure 1 can be seen to be directionally correlated to high fat containing samples compared to the lower fat samples. From Table 4, off-flavour was directionally negatively correlated to Frankfurters containing 10% and 15% fat levels and positively and significantly correlated to Frankfurters containing 20% (P<0.001) and 25% (P<0.05) fat. The higher fat content present in samples increases the propensity of lipid oxidation in the sample, which has been shown to produce a rancid and sulphur/rubber-type flavour (Byrne & Bredel, 2002). Similar findings have been shown by Faustman & Cassens (1989) and Morrissey et al. (1998).

Salt content did not have a significant effect on off-flavour perception, however, a directional correlation to higher salt levels having increased off-flavour scores can be seen in Figure 1 and Table 5. This is explained by the pro-oxidative effects of salt (Kanner, J. 1994).

The lower left quadrant in Figure 1 and Table 4 regression co-efficient indicate that frankfurtets containing 15% and 20% fat were the most acceptable fat levels to consumers, however, no significant correlations are observed. These results are in agreement with González-Viňas, Caballero, Gallego & García Ruiz (2004) who reported finding no significant differences between 10 commercially available Frankfurters as determined using a consumer panel (59 consumers) with regard to product acceptability. All the Frankfurters tested by González-Viňas et al. (2004) scored in the moderate range for overall acceptability (between 6 and 7). Healthier lipid formulation based on processing strategies is one of the most important current approaches to the development of potential meat-based functional foods.

Reformulation of Frankfurters has been used to achieve better lipid compositions by reducing fat content (Delgado-Pando et al. 2011).

Figure 1 and Table 5 show that the overall acceptability was positively and significantly correlated to the Frankfurters containing 2.5% (P<0.001) and 3.0% salt (P<0.01) and negative correlated to those containing 1.0% (P<0.001) and 1.5% (P<0.05) salt. Thus, consumers found the higher salt containing Frankfurters more acceptable than the lower salt variants. These results are not in agreement with those of Matulis et al. (1995) who produced a predictive method to determine which physico-chemical composition factors determine the acceptance of Frankfurter sausages, arriving at the conclusion that the minimum values must be 11.25% fat, 1.3% salt and a pH of 6.0

Figure 2, displays an APLSR of sensory 1 data plotted against each individual product combination of salt and fat (n=20) used in this study with the main significant correlations in this plot correlating with those found in Figure 1 and Tables 2 and 3. This plot was constructed to further investigate the interactive effects of salt and fat on the sensory properties of the Frankfurters and additional significant correlations will be reported here. Two major trends can be seen from the Figure 2. The first trend being samples containing fat levels of 10% and 15% are seen to correlate in the lower hemisphere while higher fat levels of 20% and 25% correlate in the upper hemisphere. The second major trend is that samples with higher salt levels congregate in the left side of the plot, whereas lower salt samples lie on the right side.

From Figure 2, off flavour, coarseness and toughness in are located in the very centre of the plot indicating that none of the sample scored significantly high or low in these sensory attributes. Salt perception was also shown in Figure 2 to be significantly correlated to higher salt samples and had significant negative correlations to the 10%

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fat samples (P<0.001). Again, this is due to the dilution of salt intensity as the 10% fat Frankfurters have an increased aqueous phase compared to other variants, thus diluting salt perception as the salt is dissolved in this phase. Juiciness was negatively significantly (P<0.05) correlated to 10%, 15%, and 25% fat containing samples with 1% salt as well as 10% fat samples containing 1.5% salt. This correlates with earlier findings reported in Fig. 1 and tables 2 and 3 where samples containing 1% and 1.5% salt were also significantly (P<0.001) correlated to cooking loss (Table 5). Thus these samples were perceived by consumers to be less juicy due to greater cooking losses. Meat flavour was negatively significantly (P<0.05) correlated to 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10% fat samples containing 1.5% salt. The salt acts as a flavour enhancer for natural meat flavour present in the samples and correlates with earlier findings.

Consumer acceptability is shown in the lower left quadrant to be significantly correlated to samples with lower fat and higher salt such as 15% fat with 3% salt (P<0.01), 10% fat with 3% salt (P<0.01) and 10% fat 2.5% salt (P<0.001).

6.3.2 Physiochemical analysis

In the top right quadrant of Figure 1 cooking loss can be seen to be highly significantly (Table 4, P<0.001) correlated to Frankfurters containing 10% fat. Increased cooking loss was also positively correlated (P<0.001) to lower salt levels (1.0% and 1.5% salt). Sofos (1983) found the same effect in Frankfurters and reported that a salt content in the range of 2.0–2.5% was necessary for the manufacture of commercial Frankfurters without added phosphates and in the absence of any other ingredients that might supplement the effects of sodium chloride. Additionally, Ruusunen et al. (2005) demonstrated the effect of increased salt levels on reducing cooking losses for pork and beef patties. Moisture and fat are considered to be the

major contributors to cooking loss. In terms of salt concentration effects on cooking loss we can see from Table 5 that lower salt levels, especially 1%, were significantly (P < 0.001) negatively correlated to fat content and significantly (P < 0.05) positively correlated to moisture. As expected, lower fat levels had increased moisture and decreased fat levels. Similar studies by Sampaio et al. (2004) have shown that lowering fat in Frankfurters increases cooking loss and most likely due to decreased emulsion stability.

The characteristic cured meat colour of Frankfurter is created with the use of nitrates which react with the myoglobin to form nitrosylmyoglobin before cooking and nitrosylhaemochromogen post cooking (Varnam & Sutherland, 1995). The Hunter L, a, b, values show that higher fat samples had higher L values compared to lower fat samples (10% fat), which means that they appear lighter in colour. Hunter a values, which corresponds to redness, can also be seen to correlate positively (P< 0.001) to 10% fat. The increase in lean meat explains both these findings as lean meat would be more red in colour and darker than fat. Similar results were found by Mittal & Barbut (1994), who also found that after cooking, there is far less variation in colour was found amongst samples.

Hardness, cohesiveness, chewiness and resilience, as measured by the texture analyser, were correlated significantly (P<0.05) to the higher fat samples, (20% and 25% which can clearly be seen in the regression co-efficient in Table 4 and also visible on the left of Figure 1. The variable of springiness correlated significantly (P<0.001) to Frankfurters containing 15% and 20% fat. Other research has shown that lower fat Frankfurters were harder than higher fat products (Cengiz & Gokoglu, 2007; Mittal & Barbut, 1994; Hand et al., 1987); however, contradicting results have also been reported (Candogan & Kolsarici, 2003; Hensley & Hand, 1995; Bloukas & Paneras, 1993; Gregg et al. 1993). Springiness seems to decrease for the Frankfurter containing 25% fat and this finding is in agreement with that reported by Mittal & Barbut (1994), who also found that the springiness of Frankfurters increased as fat increased, but then decreased at the highest fat levels assessed, which were between 23% and 26%.

From Figure 1 and Table 5 moisture content was significantly (P<0.001) negatively correlated to higher fat levels (20% and 25%) and positively and significantly (P<0.001) correlated to Frankfurters containing 10% fat. Additionally, Frankfurters containing 1.0% salt were positively and significantly (P<0.05) correlated to moisture content. The level of moisture is likely to have a large effect on the textural properties of the samples, specifically in relation to diluting the protein concentration. Simon et al. (1965) found that as protein content was raised, the toughness/firmness increased in beef and pork Frankfurters. Bloukas & Paneras (1993) also found hardness values positively correlated with protein content (10–14%) in low fat (10%) Frankfurters.

Figure 2, displays an APLSR of instrumental data plotted against each individual product combination of salt and fat (n=20) used in this study with the main significant correlations in this plot correlating with those found in Figure 1 and Tables 2 and 3. This plot was constructed to further investigate the interactive effects of salt and fat on the physiochemical properties of the Frankfurters and additional significant correlations will be reported here. Cooking loss was positively significantly (P<0.05) correlated to 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10% fat samples containing 1.5% and 2% salt. Hardness, Springiness, Cohesiveness, Chewiness and Resilience were all negatively significantly (P<0.05) correlated to 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10%, 15%, 20% and 25% fat containing samples with 1% salt as well as 10% fat samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well as 10% fat containing samples with 1% salt as well a

samples containing 1.5% and 2% salt. Therefore, it appears that the lower salt containing beef patties had greater cookout on cooking resulting in lower Hardness, Springiness, Cohesiveness, Chewiness and Resilience compared to the higher salt containing patties, particularly those formulated with 15%, 20% and 25% Fat.

6.4 Conclusions

Salt and fat play significant roles in the sensory and physiochemical properties of Frankfurters. Lowering the salt and fat levels is seen to have unfavourable effects on certain aspects of Frankfurter quality, such as on cooking loss and in relation to texture. Salt levels below 1.5% were shown to have a negative effect on consumer acceptability, with 2.5% salt concentrations being the most significantly preferred by consumers.

However, Frankfurters containing the lower fat levels 15% and 10% fat with higher salt levels (2.5-3%) were significantly the most acceptable variants to consumers. These results show that salt perception is very important for the consumer acceptability of Frankfurters and a reduction in levels is hard to achieve without using salt replacers. Also, fat levels can be potentially reduced without significantly affecting product quality and the overall acceptability.

6.5. Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

6.6 Tables and Figures

_	Sample	% Fat	% Pork	% Beef	% Salt	% Nitrite	% Seasoning	% Water
	F25%S3%	25.00	27.50	27.50	3.00	0.02	0.75	20.00
	F25%S2.5%	25.00	27.50	27.50	2.50	0.02	0.75	20.00
	F25%S2%	25.00	27.50	27.50	2.00	0.02	0.75	20.00
	F25%S1.5%	25.00	27.50	27.50	1.50	0.02	0.75	20.00
	F25%S1%	25.00	27.50	27.50	1.00	0.02	0.75	20.00
	F20%S3%	20.00	30.00	30.00	3.00	0.02	0.75	20.00
	F20%S2.5%	20.00	30.00	30.00	2.50	0.02	0.75	20.00
	F20%S2%	20.00	30.00	30.00	2.00	0.02	0.75	20.00
	F20%S1.5%	20.00	30.00	30.00	1.50	0.02	0.75	20.00
	F20%S1%	20.00	30.00	30.00	1.00	0.02	0.75	20.00
	F15%S3%	15.00	32.50	32.50	3.00	0.02	0.75	20.00
	F15%S2.5%	15.00	32.50	32.50	2.50	0.02	0.75	20.00
	F15%S2%	15.00	32.50	32.50	2.00	0.02	0.75	20.00
	F15%S1.5%	15.00	32.50	32.50	1.50	0.02	0.75	20.00
	F15%S1%	15.00	32.50	32.50	1.00	0.02	0.75	20.00
	F10%S3%	10.00	35.00	35.00	3.00	0.02	0.75	20.00
	F10%S2.5%	10.00	35.00	35.00	2.50	0.02	0.75	20.00
	F10%S2%	10.00	35.00	35.00	2.00	0.02	0.75	20.00
	F10%S1.5%	10.00	35.00	35.00	1.50	0.02	0.75	20.00
	F10%S1%	10.00	35.00	35.00	1.00	0.02	0.75	20.00

Sample	% Fat	% Protein	% Ash	% Moisture	% Carbohydrate
F 25% S 3%	23.15 ± 1.25	11.54 ± 0.75	3.78 ± 0.01	61.52 ± 1.23	0.01 ± 0.001
F 25% S 2.5%	25.42 ± 0.65	10.88 ± 0.62	3.46 ± 0.03	60.23 ± 1.03	0.01 ± 0.001
F 25% S 2%	23.72 ± 0.78	11.23 ± 0.51	3.01 ± 0.04	62.03 ± 0.98	0.01 ± 0.001
F 25% S 1.5%	24.00 ± 0.99	11.44 ± 0.67	3.11 ± 0.02	61.44 ± 0.74	0.01 ± 0.001
F 25% S 1%	25.41 ± 1.21	10.60 ± 0.95	2.97 ± 0.01	61.01 ± 1.03	0.01 ± 0.001
F 20% S 3%	19.18 ± 0.74	12.08 ± 0.82	4.01 ± 0.01	64.72 ± 0.88	0.01 ± 0.001
F 20% S 2.5%	20.13 ± 0.23	12.03 ± 0.63	3.35 ± 0.03	64.48 ± 0.67	0.01 ± 0.001
F 20% S 2%	19.88 ± 0.77	12.88 ± 0.42	2.68 ± 0.05	64.55 ± 0.87	0.01 ± 0.001
F 20% S 1.5%	19.63 ± 0.61	11.98 ± 0.62	2.36 ± 0.04	66.02 ± 1.14	0.01 ± 0.001
F 20% S 1%	21.27 ± 1.13	11.58 ± 0.34	2.14 ± 0.02	65.00 ± 1.07	0.01 ± 0.001
F 15% S 3%	14.68 ± 0.74	12.79 ± 0.75	4.19 ± 0.03	68.33 ± 9.70	0.01 ± 0.001
F 15% S 2.5%	14.57 ± 0.55	13.46 ± 0.64	3.33 ± 0.10	68.63 ± 0.57	0.01 ± 0.001
F 15% S 2%	15.88 ± 0.23	13.00 ± .1.01	2.99 ± 0.08	68.12 ± 0.86	0.01 ± 0.001
F 15% S 1.5%	16.29 ± 0.66	13.22 ± 0.36	2.36 ± 0.01	68.12 ± 0.49	0.01 ± 0.001
F 15% S 1%	15.98 ± 0.85	13.55 ± 0.15	2.23 ± 0.06	68.23 ± 0.92	0.01 ± 0.001
F 10% S 3%	9.61 ± 1.03	14.01 ± 0.86	4.15 ± 0.03	72.22 ± 0.76	0.01 ± 0.001
F 10% S 2.5%	10.28 ± 0.47	14.07 ± 0.46	3.55 ± 0.07	72.09 ± 0.85	0.01 ± 0.001
F 10% S 2%	10.73 ± 0.35	14.52 ± 0.61	2.87 ± 0.02	71.87 ± 1.04	0.01 ± 0.001
F 10% S 1.5%	9.93 ± 0.95	14.66 ± 0.75	2.39 ± 0.04	73.01 ± 1.23	0.01 ± 0.001
F 10% S 1%	10.75 ± 1.01	14.17 ± 0.35	1.97 ± 0.06	73.10 ± 0.70	0.01 ± 0.001

Table 2. Raw Frankfurter composition. Means values with standard deviations.

F - Fat

S - Salt

Sample	% Fat	% Protein	% Ash	% Moisture	% Carbohydrate
F 25% S 3%	$20.46 ~\pm~ 0.55$	$17.50 ~\pm~ 0.68$	$3.79 ~\pm~ 0.16$	$58.24 \ \pm \ 0.03$	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 25% S 2.5%	$20.41 \ \pm \ 0.03$	$17.72 \ \pm \ 0.45$	$3.40 \ \pm \ 0.06$	$58.46 \ \pm \ 0.13$	$0.01 ~\pm~ 0.00$
F 25% S 2%	$20.00 \ \pm \ 0.70$	$17.92 \ \pm \ 0.94$	$3.00 \ \pm \ 0.04$	59.08 ± 0.28	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 25% S 1.5%	$20.72 \ \pm \ 0.48$	$17.76 ~\pm~ 1.04$	$3.06 ~\pm~ 0.11$	$58.45 \ \pm \ 0.72$	$0.01 \hspace{0.2cm} \pm \hspace{0.2cm} 0.00$
F 25% S 1%	$20.37 \ \pm \ 0.09$	$17.67 \ \pm \ 0.95$	$3.13 ~\pm~ 0.08$	$58.81 \ \pm \ 0.10$	$0.01 \hspace{0.2cm} \pm \hspace{0.2cm} 0.00$
F 20% S 3%	$18.83 \ \pm \ 0.20$	$19.10 \ \pm \ 0.45$	$4.17 \hspace{0.2cm} \pm \hspace{0.2cm} 0.23$	$57.89 \ \pm \ 0.24$	$0.01 \hspace{0.2cm} \pm \hspace{0.2cm} 0.00$
F 20% S 2.5%	$19.14 \ \pm \ 0.03$	$18.78 \ \pm \ 0.56$	$3.39 \ \pm \ 0.07$	$58.68 \ \pm \ 0.24$	$0.01 \hspace{0.2cm} \pm \hspace{0.2cm} 0.00$
F 20% S 2%	$19.43 \ \pm \ 0.03$	$18.48 \ \pm \ 0.16$	$2.63 \ \pm \ 0.05$	$59.46 \ \pm \ 0.03$	$0.01 \hspace{0.2cm} \pm \hspace{0.2cm} 0.00$
F 20% S 1.5%	$19.61 \ \pm \ 0.36$	$18.79 \ \pm \ 1.30$	$2.45 ~\pm~ 0.09$	59.14 0.10	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 20% S 1%	$19.40 \ \pm \ 0.21$	$19.03 \ \pm \ 1.26$	$2.26 ~\pm~ 0.43$	$59.30 ~\pm~ 0.10$	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 15% S 3%	$16.27 \ \pm \ 0.46$	$19.60 \ \pm \ 0.51$	$4.09 \hspace{0.2cm} \pm \hspace{0.2cm} 0.12$	$60.03 \hspace{0.2cm} \pm \hspace{0.2cm} 0.31$	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 15% S 2.5%	$16.51 \ \pm \ 0.03$	$19.56 \ \pm \ 0.78$	$3.58 \ \pm \ 0.10$	60.34 0.18	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 15% S 2%	$16.89 \ \pm \ 0.04$	$19.34 \ \pm \ 0.94$	$3.03 \ \pm \ 0.10$	$60.73 \hspace{0.2cm} \pm \hspace{0.2cm} 0.21$	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 15% S 1.5%	$17.15 ~\pm~ 0.26$	$19.46 ~\pm~ 0.93$	$2.59 \ \pm \ 0.13$	$60.80 \hspace{0.2cm} \pm \hspace{0.2cm} 0.27$	$0.01 \hspace{0.1 in} \pm \hspace{0.1 in} 0.00$
F 15% S 1%	$17.37 \hspace{0.2cm} \pm \hspace{0.2cm} 0.08$	$19.50 \ \pm \ 0.96$	$2.13 \ \pm \ 0.11$	$60.99 ~\pm~ 0.10$	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 10% S 3%	13.67 ± 0.19	$20.36 ~\pm~ 1.49$	$3.90 \ \pm \ 0.14$	$62.06 ~\pm~ 0.40$	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 10% S 2.5%	$14.07 \hspace{0.2cm} \pm \hspace{0.2cm} 0.27$	$20.39 ~\pm~ 0.14$	$3.32 \ \pm \ 0.36$	$62.22 \ \pm \ 0.14$	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 10% S 2%	$14.04 \ \pm \ 0.17$	$20.57 \ \pm \ 0.82$	$2.76 \ \pm \ 0.12$	62.63 ± 0.07	$0.01 \hspace{.1in} \pm \hspace{.1in} 0.00$
F 10% S 1.5%	$13.44 \ \pm \ 0.32$	$20.74 \ \pm \ 0.23$	$2.34 \ \pm \ 0.13$	$63.47 \hspace{0.2cm} \pm \hspace{0.2cm} 0.16$	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F 10% S 1%	$14.61 \ \pm \ 0.22$	$21.01 \ \pm \ 1.35$	$1.94 \ \pm \ 0.15$	62.43 ± 0.29	$0.01 \hspace{0.1in} \pm \hspace{0.1in} 0.00$
F - Fat					

Table 3. Cooked Frankfurter composition. Means with standard deviations.

S - Salt

Attribute	25% Fat	20% Fat	15% Fat	10% Fat
Colour ^a	0.0027**	0.0001***	0.0625ns	-0.0001***
Texture ^a	-0.7664ns	-0.9993ns	-0.1246ns	0.1559ns
Toughness ^a	-0.2209ns	-0.0221ns	-0.0632ns	0.4783ns
Juiciness ^a	0.0054**	0.1215ns	0.0079**	-0.0461*
Salt Taste ^a	0.4453ns	0.0065**	0.0137*	-0.0001***
Meat Flavour ^a	0.2231ns	0.0001***	0.9892ns	-0.0316*
Off Flavour ^a	0.0001***	0.0088**	0.1249ns	-0.3632ns
Acceptability ^a	0.3831ns	0.7591ns	0.4736ns	-0.5795ns
Cooking Loss ^b	-0.0001***	-0.0001***	-0.1964ns	0.0001***
Cooked L Value ^b	0.5241ns	0.0001***	0.0001***	-0.0001***
Cooked a Value ^b	-0.0001***	-0.6639ns	-0.0023**	0.0001***
Cooked b Value ^b	-0.0001***	-0.0001***	-0.0148*	0.0001***
Moisture Content ^b	-0.0001***	-0.0001***	-0.8221ns	0.0001***
Fat Content ^b	0.0001***	0.0001***	0.0001***	-0.0001***
Hardness ^b	0.0001***	0.0482*	0.0001***	-0.1504ns
Springiness ^b	0.4455ns	0.0001***	0.0005***	-0.0001***
Cohesiveness ^b	0.0001***	0.0022**	0.3769ns	-0.0001***
Chewiness ^b	0.0001***	0.0001***	0.0001***	-0.2319ns
Resilience ^b	0.0001***	0.0001***	0.0654ns	-0.0001***

Table 4. Significance of estimated regression coefficients (ANOVA values) for the relationships of sensory terms ^a and instrumental ^b measurements as derived by Jackknife uncertainty testing for Frankfurters with varying fat levels.

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c P values of estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates whether the correlation is positively or negatively correlated. ^d Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01;

***=P<0.001.

Attribute	3% Salt	2.5% Salt	2% Salt	1.5% Salt	1% Salt
Colour ^a	0.2665ns	0.0296*	0.0456*	-0.8714ns	-0.0005***
Texture ^a	-0.0085**	-0.3172ns	-0.1239ns	0.0196*	0.4845ns
Toughness ^a	-0.0061**	-0.3916ns	-0.0021**	0.0018**	0.5644ns
Juiciness ^a	0.0001***	0.0001***	0.3325ns	-0.0001***	-0.0001***
Salt Taste ^a	0.0001***	0.0001***	0.9091ns	-0.0001***	-0.0001***
Meat Flavour ^a	0.0131*	0.0017**	0.1916ns	-0.0008***	-0.1542ns
Off Flavour ^a	0.9277ns	0.1528ns	0.3061ns	-0.1608ns	-0.4957ns
Acceptability ^a	0.0057**	0.0001***	0.2873ns	-0.0229*	-0.0001***
Cooking Loss ^b	-0.0001***	-0.0001***	-0.2998ns	0.0001***	0.0001***
Cooked L Value ^b	0.0018**	0.0001***	0.0092**	-0.0002***	-0.0001***
Cooked a Value ^b	-0.0001***	-0.0001***	-0.0001***	0.9923ns	0.0001***
Cooked b Value ^b	-0.0001***	-0.0001***	-0.3554ns	0.0001***	0.0001***
Moisture Content ^b	-0.7812ns	-0.0095**	-0.2378ns	0.1805ns	0.0015**
Fat Content ^b	0.8151ns	0.0141*	0.5497ns	-0.9348ns	-0.0001***
Hardness ^b	0.0001***	0.0001***	0.0001***	-0.0022**	-0.0001***
Springiness ^b	0.0001***	0.0001***	0.7115ns	-0.0001***	-0.0001***
Cohesiveness ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***
Chewiness ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***
Resilience ^b	0.0001***	0.0001***	0.0001***	-0.0001***	-0.0001***

Table 5. Significance of estimated regression coefficients (ANOVA values) for the relationships of sensory terms ^a and instrumental ^b measurements as derived by Jackknife uncertainty testing for Frankfurters with varying salt levels.

^a Sensory and hedonic terms.

^b Instrumental measurements.

^c P values of estimated regression coefficients from ANOVA-partial least squares regression (APLSR) (ANOVA values). The sign dictates weather the correlation is positively or negatively correlated. ^d Significance of regression coefficients; ns=not significant; *=P<0.05; **=P<0.01;

***=P<0.001.





Shown are the loadings of the X- and Y-variables for the first 2 PCs for \blacktriangle = the salt % and fat % = sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 42.93% and the calibrated variance was 42.21%.

Figure 2. ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for all 20 samples .



Shown are the loadings of the X- and Y-variables for the first 2 PCs for = the individual treatments, $\bullet =$ sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 47.43% and the calibrated variance was 48.91%.

Chapter 7. Effect of cooking and *in vitro* digestion on the stability of Co-Enzyme Q10 in processed meat products.

This Chapter is in the form of a manuscript submitted for publication in *Food Chemistry* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Effect of cooking and *in vitro* digestion on the stability of Co-Enzyme Q10 in processed meat products.

Abstract

The use of CoQ10 fortification in the production of a functional food has been demonstrated in the past but primarily for dairy products. This study aimed to determine the bio-accessibility of CoQ10 in processed meat products, beef patties and pork breakfast sausages, fortified with CoQ10. Both the patties and sausages were fortified with a micellarized form of CoQ10 to enhance solubility to a concentration of 1 mg/g of sample (NovaSolQ[®]). An assay was developed combining in-vitro digestion and HPLC analysis to quantify the CoQ10 present in fortified products (100 mg/g). The cooking retention level of CoQ10 in the products was found to be 74 \pm 1.42% for patties and 79.69 \pm 0.75% for sausages. The digestibility for both products ranged between 93 and 95%, sausages did have a higher digestibility level than patties but this was not found to be significant (P<0.01).

7.1 Introduction

Coenzyme Q10 (CoQ10), ubiquinone, is a ubiquitously occurring biological compound. It belongs to a homologous group of compounds called quinones which are found in many living organisms such as animals, plants and yeasts (Turunen, Olsson & Dallner, 2004). As a redox molecule, CoQ10 exists in a biologically reduced form (ubiquinol-10) and an oxidised form (ubiquinone-10) (Kubo, Fujii, Kawabe, Matsumoto, Kishida & Hosoe, 2008). Two major physiological functions have been associated with CoQ10. Namely, in mitrochondrial electron transfer within cells, producing ATP an essential component of respiration (Mitchell, 1976) and also has antioxidant activity in the reduced form (ubiquinol-10). Ubiquinone can be reduced to ubiquinol through enzymatic action post absorption (Mohr, Bowry & Stocker, 1992).

In humans, the therapeutic value of CoQ10 from supplementation in conjunction with standard medical therapy are widely recognised with respect to cardiovascular and neurodegenerative diseases (Beal, 2002; Overvad, Diamant, Holm, Holmer, Mortensen & Stender, 1999; Langsjoen & Langsjoen, 1999). Other benefits associated with CoQ10 are potentially aiding in the control of diabetes (Chew & Watts, 2004) and anti-carcinogenic properties (Lockwood, Moesgaard & Folkers, 1994). In addition, CoQ10 has been linked to delaying the onset of Parkinson's disease (Lieerman, Lyons, Levine & Myerburg, 2005 & Shults, Oakes, Kieburtz, Beal, Haas, Plumb, Juncos, Nutt, Shoulson, Carter, Kompoliti, Perlmutter, Reich, Stern, Watts, Kurlan, Molho, Harrison & Lew, 2002); and therapy post cardiac surgery to aid recovery (Rosenfeldt, Marasco, Lyon, Wowk, Sheeran, Bailey, Esmore, Davis, Pick, Rabinov, Smith, Nagley & Pepe, 2005; Rosenfeldt, Pepe, Linnane, Nagley, Rowland, Ou, Marasco & Lyon, 2002). Humans and animals biosynthesize CoQ10 which is concentrated in the heart, kidneys, liver, muscle, pancreas and thyroid gland. However, as people age the content of CoQ10 in organs decreases (Kalen, Appelkvist & Dallner, 1989). Rich sources of CoQ10 in the diets of humans are meat, poultry and fish (Stocker, 2007, Mattila, & Kumpulainen, 2001), however, the content in muscle foods is still very low (Weber, Bysted & Holmer, 1996). The total dietary intake of CoQ10 has been estimated by Kumar, Kaur, Devi & Mohan (2009) as 2-5 mg/day. This is considered too low to achieve a therapeutically beneficial effect in the body. In general the recommended therapeutic dosage is 100-200mg/day, Hathcock & Shao, (2006) reported that 2400 mg/day of CoQ10 as a maximum, with respect to tolerance and safety.

In western societies, the idea of gaining health benefits through food is becoming more and more sought after and acceptable (Kapsak, Rahavi, Childs & White, 2011). In response to this desire, a new category of food known as functional foods was developed in Japan in the 1980's. Consumers of meat products in particular are shown to be highly influenced by health and nutritional considerations (Fonseca & Salay, 2008; Angulo & Gil, 2007). The functional food market is growing at a rapid pace (Verbeke, 2005) and the future markets are optimistic (PWC, 2009; Fern, 2007). CoQ10 supplementation is commercially available as powdered filled capsules, chewable or non-chewable tablets, and soft gel capsules containing CoQ10 suspended in oil (Bhagavan, Chopra, Craft, Chitchumroonchokchai & Failla, 2007). The use of CoQ10 in the fortification of dairy foods has been successful with the use of water soluble form of CoQ10 (Pravst, Zmitek & Zmitek, 2010), however usage in the development and manufacture of a meat based functional food has not been thoroughly explored. For an ingredient to be considered useful in a functional food it requires a level of bioavailability. The bioavailability of a substance can be defined as the proportion of the ingested nutrient or active substances present in the food capable of being absorbed and available for use or storage. Bioavailability is however an imprecise concept and is difficult to measure and quantify (Ercan & El, 2011).

Bioaccessibility is another term which can be defined as the quantity of a food constituent that is present in the gut, as a result of the release of this constituent from a food medium, and maybe available to interact with an organism by intestinal absorption. Bioaccessibility includes the complete sequence of events that take place during the digestive transformation of food into material that can be assimilated by the body, the absorption/assimilation into the cells of the intestinal epithelium, and lastly, the presystemic metabolism (Fernández-García, Carvajal-Lérida & Pérez-Gálvez, 2009). Bioaccessibility is typically assessed using *in vitro* procedures (Prada & Aguilera, 2007) and is seen by many to be an excellent indicator of bioavailability.

The objective of this study is to determine the bioaccessibility of CoQ10 in processed meat products, beef patties and pork breakfast sausages, fortified with CoQ10. In this present study NovaSolQ[®] was added to both products as a source of CoQ10 to reach a concentration between 1.3 - 1.5mg/g of CoQ10 in each sample. Bioaccessibility was determined by calculating the digestibility of the CoQ10 post *in-vitro* digestion.

7.2 Materials and Methods

7.2.1 Chemicals

Chemicals purchased from Sigma Aldrich Ireland Ltd., Arklow, Co. Wicklow, Ireland, included Coenzyme Q10 standard (analytic), Hanks balanced salt solution commonly known as HBSS, lipase from porcine pancreas, sodium glycodeoxycholate, sodium taurodeoxycholate hydrate and taurocholate acid sodium hydrate, pepsin from porcine gastric mucosa, pancreatin from porcine pancreas and Methanol, 2-Propanol and Ethanol were purchased from FLUKA from LiChrosolv respectively. All reagents were certified as analytical grade. The NovaSolQ[®] was supplied by AquaNova[®] AG, Darmstadt, Germany.

7.2.2 Manufacture of Beef Patties and Pork Sausages

Beef and pork were selected on the basis of a high visual lean (V/L) score, (FSA, 2003) to ensure precise ratio of lean to fat was known. Pork shoulder was used with a V/L score of 99% and beef shin was used with a V/L score of 95%. Beef shin and pork shoulder was obtained with beef fat and pork back fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The meat and fat were vacuumpacked and stored at -18°C until required for product production.

The formulation and manufacture of beef patties was carried out as described by Tobin, O'Sullivan, Hamill & Kerry, (2012a & 2012b). To produce patties, the frozen meat and fat was then cut into strips and allowed to thaw slightly before mincing through a 5mm plate (Mincer Type: P14 TALSABELL S. A., Spain). NovaSolQ[®] was used as the source of CoQ10 for both products. The minced beef and fat was then blended together with the NovaSolQ[®] according to the formulations shown in Table 1. The respective experimental salt levels were then added and mixed
thoroughly into the beef and fat using a Stephan mixer UMC5 (Stephan U. Sohner GmbH and Co., Germany) for 45 seconds (Table 1). The mix was then weighed into portions of 100 g and formed in to patties between grease proof papers using a patty press. The patties were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored at 4°C overnight.

The formulation and manufacture of pork breakfast sausages was carried out as described by Tobin, O'Sullivan, Hamill & Kerry, (2012a & 2013). For sausage production, frozen meat and fat was cut into strips and allowed to thaw slightly before being minced through a 5mm plate (TALSABELL S. A., Spain). The meat was weighed according to the formulations shown in Table 1 and fed into the bowl chopper (Maschinenfabrik, Seydelmann, Stuttgart, D70174) with the NovaSolQ[®]. The required salt, seasoning and half the water was added and mixed at high speed for 60 seconds. The required fat was then added and the mix was chopped for further 60 seconds at high speed. The remaining water and rusk was then added and mixed at low speed for 15 seconds and high speed for 30 seconds. The sausage mix was then put into the casing filler and fed into collagen casings (DEVRO, Scotland Ltd Moodiesburn, Glasgow, G690JE). The sausages were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored in chill over night at 4°C.

7.2.3 Cooking

The beef patties and pork sausages were cooked using an oven cooking method as this was seen to be the most easily repeatable and controllable cooking method. A standard controlled cooking procedure was used for both the beef patties and pork sausages Beef patty samples were wrapped in foil and dry cooked at 150°C in a Zanussi convection oven (C. Batassi, Conegliano, Italy) for approximately 12 min

to an internal temperature of 73°C, measured using an internal temperature probe (Testo 110, Lenzkirch, Germany). Sausages were also wrapped in foil and dry cooked at 150°C for 15 minutes to an internal temperature of 73°C. Samples were cooked at the same time to insure uniformity and segregated to prevent mixing.

7.2.4 In vitro Digestion

The *in vitro* digestion model performed on the cooked beef patties and pork sausages was modelled after a method by Jiwan, Duane, O'Sullivan, O'Brien & Aherne (2010), with minor modifications. Approximately 1 g of sample was weighed and homogenized in 8 ml HBSS at 9500 rpm for 5 min using an Ultra Turrax T25 homogeniser (Janke and Kunkel, IKA-Labortechnik, GmbH and Co., Staufen, Germany). Each homogenate was then transferred to an amber bottle with 10 ml of HBSS being used to ensure the entire sample was transferred from the initial container. Porcine pepsin (0.04 g/ml HCl) was then added to the samples, followed by acidification of the samples to pH 2 using 1 N HCL thus initiating the gastric digestion process. Samples were then blanketed with nitrogen, followed by incubation at 37°C in a shaking water bath (JULABO Labortechnik GmbH SW23, Seelbach, Germany) for 1 hour.

To end the gastric phase, 0.9 M sodium bicarbonate was used to increase the pH to 5.3. After the pH was increased, 200 μ l of each bile salt was added (0.80 mM - glycodeoxycholate, 0.45 mM - taurodeoxycholate, 0.75 mM - taurocholate), as well as 100 μ l of 0.08 g/ml - pancreatin and 0.2 mg/ml - lipase. The pH was further increased to 7.4 for each sample using 0.1 M sodium hydroxide, samples were then blanketed with nitrogen and incubated at 37 °C for 2 hours in the shaking water bath to simulate the intestinal phase.

The final volume for each digested sample (digesta) was approximately 20 ml. Upon completion of the intestinal phase, 5 ml of the digesta were removed from the sample and cooled before being sealed and centrifuged at 10,000 g at 4 °C for 35 min (Beckman J2-21, Beckman Instruments Inc., CA, USA). Following centrifugation the supernatants from centrifugation were collected with a syringe and filter-sterilized using a surfactant free cellulose acetate filter (0.2 μ m; Minisart, Sartorius Stedim Biotech Minisart, Sartoriusstedim Biotech, Goettingen, Germany) and then blanketed with nitrogen gas. The samples were then stored at -80°C until required for analysis.

7.2.5 Extraction of CoQ10 form Raw, Cooked and Digested Beef Patties and Pork Sausages

Extraction of CoQ10 form raw, cooked and digested beef patties and pork sausages was performed using a modified version of the solvent extraction method described by Mattila and Kumpulainen (2001). Approximately, 1 g of raw beef and pork muscle was added to 20 ml of HBSS and homogenized with an Ultra Turrax T25 homogeniser at 9500 rpm for 5 for the extraction of initial CoQ10 content in raw sample. Undigested cooked samples were treated similarly as raw samples for the extraction of CoQ10 in cooked samples.

The digestates from 2.4 and 5 ml of homogenate for raw and cooked samples were transferred to extraction tubes, 4 ml of ethanol was added followed by homogenization at 8000 rpm for 2 min. 10 ml of n-hexane was then added to the tubes containing samples and mixed vigorously. The tubes were then centrifuged using the Beckman J2-21 centrifuge to separate the each layer at 6600 g for 5 min. The top layer of n-hexane was saved and the lower layer was re-extracted twice using 2.5 ml ethanol and 10 ml n-hexane. The collective n-hexane layers were then evaporated

(30-40 °C) using a lab-Rota C – 311 (Rescona Technics, Switzerland) and the residue was subsequently dissolved in 2.5 ml 2-propanol. The extracts were then filtered using 0.2 µm surfactant free cellulose acetate filters for HPLC analysis.

7.2.6 Coenzyme Q10 Determination

The fraction of CoQ10 present in the beef patties and pork sausages was determined by high performance liquid chromatography (HPLC) according to a method by Mattila and Kumpulainen (2001). HPLC analysis was conducted on a ProStar liquid chromatograph (Varian Analytical Instruments, CA, USA) equipped with a ProStar autosampler (Model 410, Varian Instruments) and a column oven. The sample injection volume was set at 20 µl. The column oven was set at 30 °C CoQ10 was separated on a 250 x 4 mm Nucleosil 100-5 C18 column (Macherey-Nagel GmbH & Co., Düren, Germany) and detected using a ProStar UV/Vis detector (Varian Instruments) set at 275 nm. The mobile phase consisted of methanol, 2propanol and ethanol (70:15:15, all HPLC grade) and the isocratic elution took place at a rate of 1.0 ml/min. CoQ10 was identified by comparison with the retention times of a CoQ10 standard. To prepare standard CoQ10 stock solutions 10 mg coenzyme Q10 was dissolved in 50 ml of ethanol. Working standard solutions were then prepared ranging from 2 to 200 µg/ml (A total of 7 levels of concentration; 2, 10, 40, 80, 100, 160 and 200 μ g/ml). Each CoQ10 standard solution was then filtered through a 0.2 µm surfactant free cellulose acetate filter with prior to HPLC analysis. Calibration curves were prepared by plotting peak areas versus CoQ10 concentration and regression equations were calculated; y = 11051x, $R^2 = 0.9994$. A personal computer and Star chromatography workstation software (version 5.52, Varian Inc.) was used to record the chromatograms. Standard curves were prepared for the CoQ10

and the percentage CoQ10 form sample were calculated using the calibration curves. Retention of CoQ10 post cooking was calculated as:

Retention $\% = (100/CoQ10 \text{ conc. in raw sample}) \times CoQ10 \text{ conc. in cooked sample}$

7.2.6 Statistical Analysis

All analysis was performed in triplicate and repeated in parallel to create 6 values for every sample tested (n=6). ANOVA values were calculated for the data analysed using the General Linear Model (GLM) and the statistical differences between means were measured with the post hoc Tukey test (SPSS version 20, Chicago, IL, USA). A (P < 0.01) significance level was set for all analysis, this significance level was chosen to represent the precise nature of HPLC analysis.

7.3 Results and Discussion

7.3.1 CoQ10 in Native Beef and Pork Meat and Enriched Products

All analysis of CoQ10 was completed using HPLC. An example of a chromatogram obtained from HPLC analysis of the Sigma-Aldrich Coenzyme Q10 standard (100 μ g/ml) can be seen in Figure 1. The retention time of CoQ10 calculated from the standards was 13.45 ± 0.68 min. Figure 2 shows an example chromatogram of a digested patty fortified with CoQ10 sample, it can be seen that the retention time in Figure 2 coincides with the retention time for CoQ10 in Figure 1.

Native CoQ10 concentrations in beef and pork used in this trial were found to be 48.77 \pm 4.90 µg/g and 41.64 \pm 3.31 µg/g respectively and can be seen in Table 2. No statistical significant difference (P<0.01) was found between the levels of CoQ10 in the raw beef or pork cuts used for this study (Table 2). Weber et al. (1997) reported that beef contained 31 µg/g and pork contained 14 µg/g, similar levels were found by Kubo et al. (2008) and Mattila & Kumpluainen (2001), who found beef contained 25– 36 μ g/g and pork contained 19–20 μ g/g. These results are lower than found in this present study, however reports by Pravst et al. (2010) and Kamei, Fujita, Kanbe, Sasaki, Oshipa, Otani, Matsui-Yuasa & Morisawa, (1986) showed similar higher levels for beef 16 – 40 μ g/g and pork 25- 41 μ g/g. The difference in CoQ10 level of meat has been shown to be highly dependent on the cut or organ used in analysis (Kubo et al. 2008; Mattila & Kumpluainen, 2001; Kamei et al. 1986). Other factors affecting the level of CoQ10 can include the diet of the animals, efficiency of slaughter, levels mitochondria present in the meat and freshness of the meat.

The objective of the present study was to increase the level of CoQ10 in these meats to a level where therapeutic benefits could be achieved. In general levels between 100-200 mg/day of CoQ10 is recommended to achieve a beneficial effect, however for the treatment of chronic diseases levels of up to 1200 mg/g can be used (Pravst et al. 2010; Hathcock & Shao, 2006). The level chosen was 1 mg/g (0.45% of product) for both pork sausages and beef patties under the assumption that in general the majority of the European population eats roughly between 100 and 170 g of meat in a meal (Williamson, Foster, Stanner & Buttriss, 2005). To obtain this increase NovaSolQ[®] was used as it had GRAS status and provided the CoQ10 in a clear solution and protected in an amphiphilic micelle structure. The micelles diameter was given at less than 30 nanometres and reported by the manufacturer to require no further biological micellization to pass through the small intestine.

The enriched raw sample of beef patties and pork sausages were found to contain 1358.87 \pm 34.43 µg/g and 1331.49 \pm 38.18 µg/g respectively (Table 2). The difference in concentration level is likely due to the higher level of CoQ10 present in

the source beef for the patties, however, no statistically significant (P<0.01) difference in CoQ10 concentration was observed between the two products (Table 2).

7.3.2 CoQ10 Concentration Post Cooking

The concentration of CoQ10 in the enriched beef patties and pork sausages are shown in Table 2. After cooking the beef patties were found to contain 1015.63 \pm 26.13 µg/g and the pork sausages contained 1061.03 \pm 27.70 µg/g. No significant (P<0.01) difference was found for coQ10 concentrations after cooking between the products. The retention level of CoQ10 is shown in Table 3, patties and sausages retained approximately 75% and 80% of CoQ10 respectively prior to cooking. In this study the sausages were statistically (P<0.01) found to retain more CoQ10 than the patties. This is likely due to the higher levels of cooking loss found during cooking the patties (Chapters 8 and 9). Also the collagen casings used to fill the sausage meat into provide a semi permeable barrier decreasing cooking losses.

Previous studies have reported similar loss in CoQ10 post cooking. Ercan & El (2011) found retention rates ranging from 69 to 77% in beef muscle, heart and liver when fried. Another study by Kettawan (2004) reported a 24% loss (76% retention rate) of CoQ10 after cooking. The effects of different methods of cooking on retention was reported by Ercan & El (2011) who reported decreased cooking loss of CoQ10 when samples were fried compared to boiled, postulating the loss of fat during cooking. The levels of CoQ10 destroyed by cooking in this study where samples were oven cooked seem to be most comparable to frying in terms of cooking losses.

7.3.3 CoQ10 Digestibility in Products

In Vitro digestion was carried out on the cooked beef patties and sausages; Table 3 displays the calculated level of digestibility. Digestibility is defined as the percentage of a foodstuff taken into the digestive tract that is available to be absorbed into the body. No significant (P<0.01) differences were found between the digestibility of CoQ10 in patties (93.67%) when compared to the sausages (95.06%). Similar recovery rates post digestion were found by Bhagavan et al. (2007) who reported recovery rates of approximately 98% for CoQ10 added to yogurt. The total concentration of CoQ10 in the cooked and digested beef samples was 951 ± 24.62 μ g/g (Table 2), slightly below the intended level of 1000 μ g/g. However, the concentration of CoQ10 in the cooked and digested sausages was measured to be 1008.61 ± 30.03 μ g/g. As with the cooked samples no significant (P<0.01) difference was found between the cooked and digested sausages indicate CoQ10 is very stable throughout the entire processes of cooking and digestion making it available for absorption in the intestine.

The level of CoQ10 in the beef patties after cooking compared to after cooking and digestion were found to be statistically (P<0.01) different. Statistical differences between cooked beef and cooked and digested meat CoQ10 levels here also reported by Ercan & El (2011), who found a 60% digestibility rate in fried beef muscle. The sausages in this study however did not show statistical differences between cooked and samples cooked and *in vitro* digested (Table 2). A Study by Purchas, Busboom & Wilkinson (2006) found that CoQ10 decreased after the addition of pepsin to the digestate, but later increased to levels which were not significantly different from cooked samples in beef muscle.

The absorption rate of CoQ10 is low due to its solubility in water with as low as 2–3% being absorbed in rats (Zhang, Aberg, Appelkvist, Dallner & Ernster, 1995) However, the CoQ10 added to the products in this study is in an amphiphilic micellarized form which is believed to be far more bioavailable than CoQ10 ingested from a powdered source. Bhagavan et al. (2007) assessed the uptake of CoQ10 from numerous commercially available supplements in a Caco-2 cell model system and found that CoQ10 in a micellarized form was far more readily absorbed and accumulated in the cells compared to CoQ10 in its natural non-micellarized form. Similar findings have also been reported by Chopra, Goldman, Sinatra & Bhagavan, (1998); Miles, Horn, Miles, Tang, Steele & DeGrauw, (2002) and Zaghloul, Gurley, Khan, Bhagavan, Chopra & Reddy, (2002).

From the data presented in this paper it appears that micellarized CoQ10 has a high retention rate in cooked meat systems such as patties and sausages. The addition of CoQ10 in a micellarized structure shows great promise as an additive due to the ability of the bioactive in this form not to be significantly reduced by digestion. This coupled with the low rate of cooking loss indicates that micellarized CoQ10 maintains a high degree of bioassessability when used in a meat based system.

7.4 Conclusions

The results from this study showed that the addition of micellarized CoQ10 had a higher cooking retention level and digestibility in pork sausages when compared to beef patties, likely caused by the decrease in cooking loss in sausages, however the levels were not statistically significant. These results indicate that the fortification of processed meat products with micellarized CoQ10 is achievable and can produce a product which contains as much as 79% of the CoQ10 that is present in the initial formulation before cooking. The CoQ10 was also found to have a high rate of digestibility, with as much as 95% remaining intact and reaching the site of absorption

in the small intestine. These results indicate the potential of CoQ10 as a functional ingredient in processed meat products.

7.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'. The authors acknowledge the technical assistance and advice received from both Dr. Michael O' Grady and Dr. Yvonne O'Callaghan.

7.6 Tables and Figures Table 1. Sample Formulations

	Sample Formulation								
Sample	% Beef	% Beef Fat	% Pork Fat	% Pork	% Salt	% Water	% Rusk	% Seasoning	NovaSolQ %
CoQ10 Enriched Patties	59.05	40	0	0	0.5	0	0	0	0.45
CoQ10 Enriched Sausages	0	0	22.5	42.15	1.4	20	12.5	1	0.45

Sample	CoQ10 µa/a
Raw Natural Beef (Cut - Shin)	48.77 ± 4.90^{a}
CoQ10 Enriched Patty (Raw)	1358.87 ± 34.43 ^b
CoQ10 Enriched Patty (Cooked)	1015.63 ± 26.13 ^c
CoQ10 Enriched Patty (Digested)	951.32 ± 24.62 ^d
Raw Natural Pork (Cut - Oyster)	41.64 ± 3.31 ^a
CoQ10 Enriched Sausage (Raw)	1331.49 ± 38.18 ^b
CoQ10 Enriched Sausage (Cooked)	1061.03 ± 25.70 ^c
CoQ10 Enriched Sausage (Digested)	1008.61 ± 30.03 ^{cd}

Table 2. Concentration of CoQ10 Present in Samples (µg/g)

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b, c, d means with different letters in columns are significantly different (p < 0.05). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.

Sample	Post Cooking Retention %	Digestability %
Beef Patties	74.74 ± 1.42 ^a	93.67 ± 1.85 ^a
Sausages	79.69 ± 0.75 ^b	95.06 ± 1.86 ^a

Table 3. Retention of CoQ10 in Samples Post Cooking and Digestability of CoQ10

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b means with different letters in columns are significantly different (p < 0.01). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.

Figure 1. Chromatogram of CoQ10 Standard 100 $\mu g/ml$ (mAu versus Minutes). The retention time for CoQ10 was calculated at 13.45 ± 0.68



Figure 2. Chromatogram of Digested Enriched Patty (mAu versus Minutes). Peak corresponds with CoQ10 retention time of CoQ10 peaks from Sigma-Aldrich standard (Figure 1).



Chapter 8. Consumer Evaluation of the Commercial Viability of Reduced Salt and Fat Beef Patties and Patties Fortified with Co-Enzyme Q10.

This Chapter is in the form of a manuscript submitted for publication in *Meat Science* as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Consumer Evaluation of the Commercial Viability of Reduced Salt and Fat Beef Patties and Patties Fortified with Co-Enzyme Q10.

Abstract

Functional foods are growing in popularity with consumers. This has occurred through the increasing association of specially formulated consumer foods to wellbeing. The functional food sector within the meat industry is beginning to evolve. The aim of this study was to assess consumer attitudes to functionally formulated beef patties possessing reduced salt and fat levels and enriched with CoQ10 when compared against current conventional beef patty products available commercially in the retail market.

Reduced fat and salt patties (40% fat; 0.5% salt), with and without the addition of CoQ10 (100 mg/g sample), were compared by consumer sensory evaluation (n=100) against commercially available products. No significant differences were found between the reduced salt and fat products compared to products fortified with CoQ10. The reduced fat and salt, as well as the CoQ10 fortified patties were more accepted by consumers compared to the commercially available products and scored significantly (P < 0.05) higher for appearance.

8.1 Introduction

Studies conducted around the globe have shown that consumers believe that the foods they consume directly impact on their health and well-being (Mollet & Rowland, 2002; Young, 2000). Health concerns over meat consumption are especially important for consumers (Fonseca & Salay, 2008; Angulo & Gil, 2007). More recent studies have shown that consumers view processed meats, in particular, as an unhealthy product (Chapter 3).

Meat is associated with both positive and negative nutritional attributes. Meat in itself is an excellent source of high, biologically valuable protein and contains both vitamins and minerals. Fresh meat is naturally low in sodium, and contains multiple endogenous antioxidants and other bioactives. (Williams, 2007; Jimenez-Colmenero, Carballo & Cofrades, 2001). Many consumers have developed a negative perception especially over nutritional qualities of meat and meat products, these concerns include; high levels of saturated fat; cholesterol; high levels of sodium and other harmful chemicals (Chapter 3; Whitney & Rolfes, 2002).

To reduce concerns over health a great deal of research has been conducted to reduce or replace fat, sodium and nitrite levels in processed meats (Tobin, O'Sullivan, Hamill, & Kerry, 2012ab; Tobin, O'Sullivan, Hamill, & Kerry, 2013; Viuda-Martos, Fernández-López, Sayas-Barbera, Sendra, Navarro, & Pérez-Álvarez, 2009; Yang, Choi, Jeon, Park, & Joo, 2007; Ruusunen, Vainionpaa, Lyly, Lahteenmaki, Niemisto & Ahvenainen, 2005; Dineen, Kerry, Lynch, Buckley, Morrissey, & Arendt, 2000; Solheim & Ellekjær, 1993; Maurer, 1983). With these new formulations the industry strives to change consumers' beliefs and attitudes regarding processed meats to those which are more informed, realistic and balanced. Making meat a more acceptable product to consumers is essential for the meat industry to develop commercially successful functional foods. The functional food industry is a rapidly growing sector of the food industry and is estimated to be worth \$130 billion by 2015 (Global Industry Analysts, 2010). A Functional food is a product that is derived from naturally-occurring ingredients; consumed daily as part of an overall diet and provides health benefits beyond basic nutrition (Jimenez-Colmenero et al. 2001).

Numerous strategies can be implemented to create meat-based functional foods ranging from dietary supplementation of the animal to addition of a functional ingredient to an existing product. Co-enzyme Q10 (CoQ10) is an example of a functional bioactive that could be incorporated into processed meat products. CoQ10 has principally been linked to aid and offer protection from cardiovascular and neurodegenerative diseases (Beal, 2002; Overvad, Diamant, Holm, Holmer, Mortensen & Stender, 1999; Langsjoen & Langsjoen, 1999). Other benefits associated with CoQ10 include; diabetes control (Chew & Watts, 2004); anti-cancer (Lockwood, Moesgaard & Folkers, 1994); Parkinson's disease (Shults, Oakes, Kieburtz, Beal, Haas, Plumb, Juncos, Nutt, Shoulson, Carter, Kompoliti, Perlmutter, Reich, Stern, Watts, Kurlan, Molho, Harrison & Lew, 2002); or as an adjunct therapy after cardiac surgery (Rosenfeldt, Marasco, Lyon, Wowk, Sheeran, Bailey, Esmore, Davis, Pick, Rabinov, Smith, Nagley & Pepe, 2005; Rosenfeldt, Pepe, Linnane, Nagley, Rowland, Ou, Marasco & Lyon, 2002)

The dietary intake of CoQ10 from food is 2-5 mg/day (Kumar, Kaur, Devi & Mohan, 2009). However, this is to too low to achieve any beneficial effect in the body. Therapeutic doses are generally recommended as 100-200 mg/day. Therefore, CoQ10 is considered by some nutritionists to be suitable as a dietary supplement.

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However, creating a functional food is a difficult task as products must be produced without dramatically affecting the consumers need for quality, convenience and price.

The objective of this study is to compare the consumer sensory evaluation of reduced salt and fat beef patties to commercially available patties, and also to investigate consumer sensory acceptance of beef patties enriched with CoQ10.

8.2 Materials and Methods

8.2.1 Sample Preparation

Beef was selected on the basis of a high visual lean (V/L) score, (FSA, 2003); beef shin was used with a V/L score of 95%. Beef was purchased along with beef fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The meat and fat were then vacuum-packed and stored at -18°C until required for product production. A band saw was used to cut the frozen meat and fat into strips and which were then allowed to thaw slightly before being minced through a mincer (Mincer Type: P14 TALSABELL S. A., Spain). NovaSolQ[®] (AquaNova[®] AG, Darmstadt, Germany) was used as the source of CoQ10 for the CoQ10 enriched patties. The minced beef and fat were then blended together with the NovaSolO[®] to achieve a level of 100 mg/100 g of patty (1000 ppm), for samples not containing NovaSolQ[®] this step was carried out to blend together the fat and meat. The salt level (0.5%) was then added and mixed thoroughly into the meat and fat (40%) by a Stephan mixer UMC5 (Stephan U. Sohner GmbH and Co., Germany) for 45 seconds (Table 1). Patties were then formed using approximately 100 g of the mix in a patty press between grease proof papers. The patties were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored at 4°C overnight.

8.2.2 NovaSol $Q^{\mathbb{B}}$

To produce CoQ10 fortified patties NovaSolQ[®] (AquaNova[®] AG, Darmstadt, Germany) was used as it had GRAS status. NovaSolQ[®] also provides the CoQ10 in a clear solution and using an amphiphilic micelle structure aids in protecting the CoQ10 from environmental stresses. The micelles diameter was given at less than 30 nonometers and reported by the manufacturer to require no further biological micellization to pass through the small intestine.

8.2.3 Commercial Products

Two-commercially available leading brands of beef patties were bought from supermarkets found throughout Ireland. Compositional testing was carried out on each product in triplicate; the composition of these products is shown in Table 1. Commercial Patty 1 was purchased on the basis that it was a higher quality product and Commercial Patty 2 was chosen as it represented lower cost product.

8.2.4 Cooking

Oven-cooking was the chosen cooking method used in this study as it provided greater control and was more repeatable than other options. All patty samples (including commercial brands) were wrapped in foil and dry-cooked at 150°C in a Zanussi convection oven (C. Batassi, Conegliano, Italy) for approximately 12 minutes to an internal temperature of 73°C, as measured by an internal temperature probe (Testo 110, Lenzkirch, Germany).

8.2.5 Sensory Evaluation

Sensory analysis was carried out using 100 consumers within the age range 18–65 years. Panellists were chosen on the basis that they regularly consume and purchase burger type meat products. Sensory analysis was undertaken in the panel booths at the university sensory laboratory that conforms to ISO (1988) international standard. Consumers were presented with four samples on paper plates. The consumers were directed to rinse their mouths with water before tasting each sample. Sample presentation order was randomised to prevent any flavour carryover effects (MacFie, Bratchell, Greenhoff & Vallis, 1989). The consumers were then instructed to indicate their score on a 10 cm line scale ranging from 0 at the left to 10 at the right and rating subsequently scored in cm from left for each sample presented (O'Sullivan, Byrne & Martens, 2003). Consumers were required to evaluate every sample using the following descriptors: appearance, texture, tenderness, juiciness, salt taste, meat flavour, fat flavour, overall flavour intensity, off-flavour, oxidative flavour and overall acceptability. Oxidative flavour was described to consumers as rancid, cardboard or linseed oil-like flavour.

8.2.6 Protein Content

In order to measure protein concentrations the Kjeldahl method (Suhre, Corrao, Glover and Malanoski, 1982) was used. A digestion block was pre-heated to 410°C. Approximately 0.5 g of well homogenised sample was weighed accurately into a digestion tube. 15ml of sulphuric acid (nitrogen free), 10 ml hydrogen peroxide and 2 "kjeltabs" were added to the sample in the tube. The tubes where then placed into the pre-heated digestion block. Upon the samples becoming colourless they were

removed from the block. The tubes were allowed to cool in the fume hood after removal.

Inside the fume-hood, 50 ml of distilled water was carefully added to the cooled and digested sample. The tubes and a receiver flask containing 50 ml of 4% Boric acid, with indicator, were then inserted into the distillation unit. After the sample had been distilled the contents of the receiver flask were titrated against 0.1 N hydrochloric acid until the green colour reverted back to the original red colour.

8.2.7 Ash Content

Ash content was determined with the use of a muffle furnace (AOAC, 1923). The muffle furnace was firstly pre-heated to 525°C. Approximately 5 g of well homogenised sample was weighed into a porcelain dish using a balance that weighs to 1 mg. The dish containing the sample was then put in the muffle furnace until the colour of the samples went white (approximately 6 hours). The dishes containing the samples were then removed and placed in desiccators to cool. The dishes were then weighed and the ash content calculated.

8.2.8 Moisture and Fat Content

A total of 200 g of sample was homogenised in a Büchi Mixer B-400 (BÜCHI Labortechnik AG, Meierseggstrasse 40, Postfach, CH-9230 Flawil 1, Switzerland). The homogenised sample was then quickly transferred into a moisture proof bag to avoid moisture or evaporative loss. Moisture and fat content were then determined using the CEM SMART (moisture) and SMART Trac (fat) systems (Bostian, Fish, Webb, and Arey, 1985). Both raw and cooked samples were measured for colour. The surface of the patties were measured for colour according to the CIE L* a* b* colour system, Cooked samples were cooled to room temperature before measuring. A Minolta CR 300 colorimeter (Minolta Camera Co. Ltd., Osaka, Japan) with an 11 mm-diameter aperture, D_{65} illuminant, calibrated by the CIE Lab colour space system using a white tile (C: Y = 93.6, x = 0.3130, y = 0.3193), (Minolta calibration plate) was used to conduct all analysis. Colour measurements (CIE L^* , a^* and b^* values representing lightness, redness and yellowness, respectively) were taken. Nine readings were taken per sample.

8.2.10 Cooking Loss

Patties sample weights were recorded before and after cooking and the differences in weights recorded. The samples were cooked as described in 2.2. Before weighing, samples were blotted with a paper towel to remove excess surface moisture.

Calculation for cook loss was as follows:

% cook loss = ((cooked weight – raw weight)/raw weight) x 100

8.2.11 Texture Analysis

Texture profile analysis (TPA) was used to obtain texture measurements for individual samples. All analysis was performed at room temperature with a Texture Analyser 16 TA-XT2i (Stable Micro Systems, Surrey, UK). Samples were subjected to a two-cycle compression test using the 25 kg load cell. The samples were compressed to 40% of their original height with a cylindrical probe (SMSP/100 Compression plate) 100 mm diameter and a cross-head speed of 1.5 mm/sec.

Texture profile parameters were determined following descriptions by Bourne (1978) and the SMS manual (Stable Micro Systems, Surrey, UK), these included; hardness (N) maximum force required for the initial compression of the sample; springiness (mm), the samples ability to recover its original shape after the initial compression and the deforming force was removed; adhesiveness ($N \times mm$), area under the abscissa post initial compression; cohesiveness (Dimensionless), extent to which the sample could be deformed prior to rupture, measured by the areas under the compression portion only and excludes the areas under the decompression portion instead of using the total area under positive force; chewiness ($N \times mm$), the required work to masticate the sample, measured as the product of hardness times cohesiveness times springiness; and resilience (Dimensionless), the ratio between the negative force input to positive force input during the first compression.

8.2.12 Salt Determination

The Volhard method for salt determination (AOAC, 1995) was used to determine the salt levels for each sample. Between 2.5 and 3 g of sample was weighed into each conical flask. 25 ml of 0.1 N Silver Nitrite solution and 15 ml of concentrated Nitric acid was added. Sufficient boiling chips were added to the flasks and the samples were boiled until the meat digested. Potassium Permanganate was added in small doses while boiling until the solutions maintained the dark colour for several minutes before reverting back to clear. 25 ml of distilled water was then added and the solutions were allowed to boil for approximately 5 minutes. before being removed from the heat source and allowed to cool to room temperature. Once at room

temperature the samples were diluted to approximately 150 ml with distilled water. 2 ml of ferric alum indicator was then added to each sample and excess Silver Nitrite was titrated against Potassium Thiocyanate.

8.2.13 Data Analysis

The data accumulated from the 100 test subjects during the sensory evaluation and data acquired by instrumental methods was processed using ANOVA-partial least squares regression (APLSR). APLSR, is based on cross-validation and stability plots (Martens and Martens, 1999, 2001). Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway) was used to perform all analyses. The X-matrix was designed as 0/1 design variables for the patties. The sensory, chemical, and instrumental variables were used for the Y-matrix design.

The optimal number of components in the APLSR models presented was determined to be four principal components; PC 1 versus PC 2 is presented in Figure 1; other PCs did not yield any more additional information or provide any predictive improvement in the Y-matrix obtained prior examination. The validated explained variance for the model constructed was 32.70% and the calibrated variance was 32.05% for Figure 1.

The significant effects of consumer sensory evaluation were carried out on the raw sensory data and analyzed using the General Linear Model (GLM) with post hoc Duncan comparison of mean scores (SPSS, Chicago, IL, USA). Significance level was set at (P < 0.05) for all analysis. These values are presented in Table 2 for sensory variables and Table 3 for physiochemical variables.

8.3 Results and Discussion

8.3.1 Sample Preparation

The results from the consumer sensory evaluation which was carried out by 100 regular consumers of processed meats, and the instrumental data figures for each product type, are presented in the APLSR plots in Figure 1. Mean values for each sensory variable are shown in Table 2, and for physiochemical data in Table 3.

Formulations for the non-commercial patties were determined from previous work by Tobin et al. (2012a). Consumer sensory analysis carried out by Tobin et al. (2012b) found that lower fat and salt patties were significantly preferred by the consumer. NovaSolQ[®] was used in the formulation of the CoQ10 enriched products using the basic formulations from Tobin et al. 2012a.

Compositional analysis was carried out on each sample after cooking and is displayed in Table 1. The primary losses during cooking in meat products are fat and moisture. It can be seen that the reduced salt and fat patty (RP) and the CoQ10 fortified patty (CoP) had lower (P<0.05) fat levels than the commercial products (Table 1) and also higher moisture levels (Table 1).

Cooking loss was found to be highly correlated to RP and CoP and when compared to commercial patties 1 (CP1) and 2 (CP2). Salt levels, play a significant role in cooking loss, Tobin et al. (2012a) and Ruusunen et al. (2005) have shown the effect of increased salt levels on reducing cooking losses from meat patties. Salt, is used by industry to solubilise myofibrillar protein and consequently to assist in the binding of water and in the encapsulation of fat within the matrix of the meat products.

Higher levels of moisture were detected in the reduced salt and fat products as well as in the CoQ10 fortified products (Table 1). These findings are as a direct result of higher levels of lean meat being present within these products. Lean meat contains far more water than fat and also possesses quite high levels of myofibrilar protein which is available to bind to water following the addition of salt (Foegeding & Lanier, 1987; Acton & Dick, 1984). The increase in lean meat content also accounts for the higher protein levels determined in RP and CoP treatments.

8.3.2 Sensory Analysis

All sensory analysis was conducted on cooked patties. Fig. 1 displays the ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot of sensory and instrumental analysis for experimental and commercial beef patties Appearance of the cooked patties, as shown in Figure 1, was found to be the most correlated to CP1, CoP and the RP. However in Table 2 it is shown that the CoP and the RP scored significantly (P<0.05) higher in liking of appearance than the two commercial patties. Consumer purchases of meat are primarily based on visual sensory evaluation (O'Sullivan et al. 2002) and appearance is a very important meat quality factor and the appearance of a bright red colour indicates freshness and wholesomeness (Zakrys-Waliwander, O'Sullivan, Walshe, Allen & Kerry, 2011). Colour measurements using the CIE L* a* b* system showed that the RP and CoP were lighter (P<0.05) in colour than (L* value) and also more red (a* value) than the less accepted commercial sample CP2 (Table 3). Hughes, Cofrades & Troy (1997) reported that fat affects the colour parameters of cooked meat products. Research carried out by Tobin et al. (2012a) showed that patties with higher colour scores, in terms of lightness and redness, were found to be most acceptable to the consumer. No significant difference was found between consumer scoring of the RP and CoP, which showed that the addition of CoQ10 had no negative impact on product appearance. CIE L* a* b* value, did however, display significant (P<0.05) differences between b*

values (Yellowness/Blueness), CoP was found to be significantly (P<0.05) more yellow when compared to the RP treatment. This can be attributed to the yellow colour associated with CoQ10. Interestingly, the instrumental detection of a yellow hue in meat was not perceived by consumers and consequently, had no negative impact on consumer acceptance. This lack of correlation between instrumental colour analysis and consumer panels was also observed by this research group for a range of other sausage type meat products (Chapter 9).

The overall scores for liking of texture was positively correlated to the CoP, RP and CP1 as can be seen in the bottom hemisphere of Figure 1 and negatively correlated to CP2. None of the three patties, which positively correlated to texture, scored significantly higher than each other. However, CP2 was found to score significantly (P<0.05) lower for liking of texture than the other products (Table 2). The most tender product found by panellists was CP1 (Table 2) and was found to be significantly (P<0.05) more tender than CP2, but not significantly different to RP and CoP treatments. The decrease in consumer tenderness ratings for CP2 can be explained by its higher salt to protein ratio compared to CP1, reduction in salt has been shown to decrease toughness because of a decrease in the solubilisation of myofibrillar protein (Schwartz & Mandigo, 1976). No significant differences were found between CoP and RP.

Texture profile analysis of the patties showed that the RP and CoP were less (P<0.05) hard than the commercial patties Table 3. The higher salt levels in the CP1 and CP2 would increase the solubilisation of the functional myofibrillar proteins, therefore producing a tougher product resulting in an increase in hardness (Acton & Dick, 1984). RP and CoP treatments had higher (P<0.05) springiness scores than those observed for commercial patties (Table 3). The increase in myofibrillar gel

network due to increased salt increases the likeliness of fracturability which decreases the springiness of the product. RP and CoP were less (P<0.05) chewy than both commercial patties. Chewiness is defined as the effort required to masticate a sample, the lower the chewiness score the easier it is to chew. Both product tenderness and texture play a vital role on the overall chewiness of a product. The RP and CoP (as shown in Figure 1) have been shown to have a preferential texture by consumer sensory analysis over the commercial patties and which correlates with chewiness values.

Consumers rated CP1 as being the juiciest of all the patties assessed; (Figure 1). Table 2 shows that CP1 was significantly (P<0.05) juicier than all other patties. These results are in agreement with previous research carried out by Berry & Leddy (1984) and Tobin et al, (2012a) who found that ratings for juiciness in higher fat patties were higher than that of lower fat patties. No significant differences were found between CoP and RP. However, CoP was shown to be significantly juicier than CB2 (Table 2).

The saltiest tasting patties were found to be CP1, and the least salty was CP2. The decrease in perceived saltiness in CP2 is likely caused by the addition of spices which mask the taste of salt in the product. The addition of CoQ10 was found to have no impact on consumer salt perception of patties. CP2 was also found to have a less significant (P<0.05) (Table 2) meat flavour than the other three patties, with no significant differences being found between the other samples. Fat flavour was highest in CP1, which was significantly (P<0.05) higher than that detected for CoP, which was reported as having the least level of fat flavour.

In terms of overall flavour intensity RP, CoP and CP1 did not differ statistically, however CP2 had lower (P<0.05) overall flavour intensity. CP2 was

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more (P<0.05) positively correlated to Off-Flavour than CoP, but not statistically more than the other two products. None of the products were found to be more statistically correlated to Oxidative Flavour. The RP, CoP and CP1 were more (P < 0.05) acceptable than CP2. RP and CoP scored higher for overall acceptability than CP1, but this difference was not statistically significant.

8.4 Conclusion

Reduced fat and salt patties and those enriched with CoQ10 were found to have a better appearance compared to commercial products, but performed similarly to CB1 in terms of tenderness, juiciness and flavour. The addition of CoQ10 into patties was found to have few if any impact on consumer sensory acceptance. The most significant change to samples was found to be an increase in yellowness (b* value), which was not perceived by consumers. In conclusion, the reduction of fat and salt in the patties produced products which consumers found to be more acceptable than commercially available products. Similarly the reduced salt/fat patties fortified with CoQ10 were also found to be more acceptable than commercially available products.

8.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

8.6 Tables and Figures: Table 1. Compositional analysis

	Code	Moisture	Fat	Protein	Carbohydrate	Salt
Burger 40%Fat 0.5%Salt	RP	59.76 \pm 0.10 ^a	14.05 ± 0.22^{a}	24.80 ± 0.37^{a}	0.01 ± 0.01^{a}	0.50 ± 0.01^{a}
Burger CoQ10 (1000 ppm)	CoP	57.00 ± 0.04^{b}	14.87 ± 0.27^{b}	26.71 ± 0.23^{b}	$0.01~\pm~0.01^a$	0.51 \pm 0.01 a
Commercial Patty 1	CP1	54.65 ± 0.09^{c}	21.15 ± 0.22^{c}	21.60 ± 0.06^{c}	1.91 ± 0.01^{b}	$0.70 ~\pm~ 0.01^b$
Commercial Patty 2	CP2	52.02 ± 1.09^{d}	$22.88 \pm 0.08^{\text{d}}$	$23.11 \pm 0.69^{\circ}$	$2.21 \pm 0.01^{\circ}$	$0.84 \pm 0.01^{\circ}$

	Appearance	Texture	Tenderness	Juiciness	Salt Taste	Meat Flavour	Fat Flavour	Overall Flavour	Off Flavour	Oxidative Flavour	Overall Acceptability
								interiory		Thateal	71000ptability
Burger 40%Fat 0.5%Salt	6.92 ± 0.16^{a}	6.39 ± 0.22^{a}	6.44 ± 0.18^{ab}	4.89 ± 0.22^{ac}	4.07 ± 0.19^{a}	6.77 ± 0.18^{a}	5.68 ± 0.21^{ab}	6.30 ± 0.20^{a}	3.33 ± 0.21^{ab}	3.74 ± 0.21^{a}	6.58 ± 0.20^{a}
Burger CoQ10	$6.62 \ \pm \ 0.19^{a}$	$6.45 \ \pm \ 0.20^{a}$	$6.61~\pm~0.17^{ab}$	$5.31 \ \pm \ 0.19^{a}$	$4.29 \ \pm \ 0.18^{a}$	$6.39 \ \pm \ 0.19^{a}$	$4.91 \ \pm \ 0.20^{a}$	$6.23 ~\pm~ 0.19^{a}$	$3.19 \ \pm \ 0.20^{a}$	3.71 ± 0.21^{a}	$6.56 \ \pm \ 0.19^{a}$
Commercial Burger 1	5.31 ± 0.26^{b}	6.08 ± 0.25^{a}	7.09 ± 0.18^{a}	6.97 ± 0.21^{b}	$5.36 ~\pm~ 0.22^{b}$	6.88 ± 0.20^{a}	5.78 ± 0.21^{b}	6.60 ± 0.24^{a}	$3.51 \ \pm \ 0.23^{ab}$	3.41 ± 0.21^{a}	6.49 ± 0.23^{a}
Commercial Burger 2	$4.78 \pm 0.22^{\texttt{b}}$	$4.85 \pm 0.23^{\texttt{b}}$	$6.06~\pm~0.20^{\textrm{b}}$	4.45 ± 0.22^{c}	3.67 ± 0.18^{a}	$5.13 \pm 0.25^{\text{b}}$	$5.05~\pm~0.23^{\text{ab}}$	$4.59~\pm~0.23^{\textrm{b}}$	$4.01 \hspace{0.1in} \pm \hspace{0.1in} 0.25^{\texttt{b}}$	3.70 ± 0.21^{a}	$4.78 \pm 0.23^{\texttt{b}}$

Table 2. Sensory Mean Values and Standard Deviation

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b, c, d means with different letters in columns are significantly different (p < 0.05). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.

	Cooking Loss	Raw L value	Raw a value	Raw b value	Cooked L value	Cooked a value	Cooked b value	Cooked Salt
Reduced Salt/Fat Burger Burger CoQ10	0.40 ± 0.01^{a} 0.46 + 0.01 ^b	54.53 ± 3.97^{a} 51.83 + 1.25 ^b	18.33 ± 2.06^{a} 21.23 + 0.76 ^b	11.64 ± 1.04^{a} 19.02 + 0.38 ^b	48.58 ± 2.15^{a} 46.57 ± 0.78^{b}	9.99 ± 0.44^{a} 9.99 ± 0.38^{a}	8.16 ± 0.53^{a} 12 23 + 0.20 ^b	0.50 ± 0.01^{a} 0.51 + 0.01 ^a
Commercial Burger 1 Commercial Burger 2	$\begin{array}{rrrr} 0.35 \pm & 0.02^{c} \\ 0.17 \pm & 0.02^{d} \end{array}$	$47.54 \pm 0.71^{\circ}$ $60.20 \pm 1.36^{\circ}$	$\begin{array}{r} 22.65 \pm 0.59^{\circ} \\ 15.29 \pm 1.49^{\circ} \end{array}$	$15.70 \pm 0.42^{\circ}$ $15.23 \pm 1.14^{\circ}$	$\begin{array}{r} 42.02 \pm 1.49^{c} \\ 39.39 \pm 2.36^{d} \end{array}$	9.37 ± 0.24^{b} 9.79 ± 0.21^{c}	$14.92 \pm 2.30^{\circ}$ $11.54 \pm 0.35^{\circ}$	0.70 ± 0.01^{b} 0.84 ± 0.01^{c}
	Cooked Moisture	Cooked Fat	Cooked Protein	Hardness	Springiness	Cohesiveness	Chewiness	Resilience
Reduced Salt/Fat Burger	Cooked Moisture 59.76 \pm 0.10 ^a	Cooked Fat 14.05 \pm 0.22 ^a	Cooked Protein 24.80 ± 0.37^{a}	Hardness 17535.75 ± 513.48 ^a	Springiness 1.29 ± 0.47 ^a	Cohesiveness 0.61 ± 0.01^{a}	Chewiness 13883.00 ± 554.11 ^a	Resilience 0.50 ± 0.01^{a}
Reduced Salt/Fat Burger Burger CoQ10	$\begin{array}{r} \mbox{Cooked Moisture} \\ 59.76 ~\pm~ 0.10^{a} \\ 57.00 ~\pm~ 0.04^{b} \end{array}$	$\begin{array}{rl} \mbox{Cooked Fat} & & \\ 14.05 \pm 0.22^a & \\ 14.87 \pm 0.27^b & \end{array}$	$\begin{array}{r} \mbox{Cooked Protein} \\ \mbox{24.80} \pm \ 0.37^{a} \\ \mbox{26.71} \pm \ 0.23^{b} \end{array}$	Hardness 17535.75 ± 513.48^{a} 17858.32 ± 482.16^{a}	Springiness 1.29 ± 0.47 ^a 1.21 ± 0.47a	$\begin{array}{r} \text{Cohesiveness}\\ 0.61~\pm~0.01^a\\ 0.62~\pm~0.04^b \end{array}$	$\begin{array}{r} \mbox{Chewiness} \\ 13883.00 ~\pm~ 554.11^a \\ 13274.63 ~\pm~ 644.46^a \end{array}$	$\begin{array}{r} \text{Resilience} \\ 0.50 ~{\pm}~~ 0.01^{a} \\ 0.53 ~{\pm}~~ 0.04^{b} \end{array}$
Reduced Salt/Fat Burger Burger CoQ10 Commercial Burger 1	$\begin{array}{rl} \mbox{Cooked Moisture} \\ \mbox{59.76} \pm \ 0.10^{a} \\ \mbox{57.00} \pm \ 0.04^{b} \\ \mbox{54.65} \pm \ 0.09^{c} \end{array}$	$\begin{array}{r} \mbox{Cooked Fat} \\ 14.05 ~\pm~ 0.22^a \\ 14.87 ~\pm~ 0.27^b \\ 21.15 ~\pm~ 0.22^c \end{array}$	$\begin{array}{r} \mbox{Cooked Protein} \\ \mbox{24.80 } \pm \ 0.37^a \\ \mbox{26.71 } \pm \ 0.23^b \\ \mbox{21.60 } \pm \ 0.06^c \end{array}$	$\begin{array}{r} \mbox{Hardness} \\ 17535.75 \pm 513.48^a \\ 17858.32 \pm 482.16^a \\ 23360.28 \pm 684.16^b \end{array}$	$\begin{array}{r} \text{Springiness} \\ 1.29 ~\pm~ 0.47^a \\ 1.21 ~\pm~ 0.47a \\ 0.91 ~\pm~ 0.01^b \end{array}$	$\begin{array}{r} \text{Cohesiveness} \\ 0.61 \ \pm \ 0.01^a \\ 0.62 \ \pm \ 0.04^b \\ 0.58 \ \pm \ 0.01^c \end{array}$	$\begin{array}{r} \mbox{Chewiness} \\ 13883.00 ~\pm~ 554.11^a \\ 13274.63 ~\pm~ 644.46^a \\ 14325.78 ~\pm~ 169.88^b \end{array}$	$\begin{array}{r} \text{Resilience} \\ 0.50 \ \pm \ 0.01^a \\ 0.53 \ \pm \ 0.04^b \\ 0.51 \ \pm \ 0.01^a \end{array}$

Table 3. Physio-chemical Analysis Mean Values and Standard Deviation

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b, c, d means with different letters in columns are significantly different (p < 0.05). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.



Figure 1. ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for patties.

Shown are the loadings of the X- and Y-variables for the first 2 PCs for = the individual treatments, $\bullet =$ sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 32.70% and the calibrated variance was 32.05%.

Chapter 9. Consumer Evaluation of Reduced Salt and Fat Sausages and Frankfurters and Variants Fortified with Co-Enzyme Q10.

This Chapter is in the form of a manuscript submitted for publication in LWT as follows:

Tobin, B., O'Sullivan, M. G., Hamill, R. & Kerry, J. P. (2013). Consumer Evaluation of Reduced Salt and Fat Sausages and Frankfurters and Variants Fortified with Co-Enzyme Q10.
Abstract

The functional food sector is growing as health and nutritional concerns become more important to consumers. This growth is driving the development of healthier food products, with the meat industry being no exception. The objective of this study was to assess consumer attitudes to functionally formulated sausages and Frankfurters possessing reduced salt and fat levels and enriched with CoQ10 when compared against current conventional sausage-style products available commercially in current markets in the UK and Ireland.

Reduced fat sausages (22.5% Fat; 1.4% Salt) and Frankfurters (10% Fat; 2.5% Salt), formulated both with and without the addition of CoQ10 (100 mg/g meat), were compared by consumer sensory evaluation against commercially available products (2 sausage and 2 Frankfurter formats). No significant differences were determined between the reduced salt and fat products compared to products fortified with CoQ10. The reduced fat and salt, as well as the CoQ10 fortified, sausages were found to compare quite well to their commercial counterparts for overall acceptability, whereas commercial Frankfurters were found to be the more favoured in comparison to reduced fat and CoQ10 fortified Frankfurters.

9.1 Introduction

With heart disease and other chronic illnesses affecting an ever-expanding percentage of the population, consumers are progressively becoming more aware of health issues and preventative measures they can adopt to reduce their chances of becoming ill. In more recent years, global scientific studies have shown that consumers place a greater importance in the health and nutritional value of the foods they consume (Mollet & Rowland, 2002; Young, 2000). Over the past decades health scares within the meat industry have damaged public perception (Coffey, Mintert, Fox, Schroeder & Valentin, 2005; Marsh, Schroeder & Mintert, 2004). Such negativity creates greater concern over meat consumption by the general public (Fonseca & Salay, 2008; Angulo & Gil, 2007). Tobin, O'Sullivan, Hamill & Kerry (2012a) have shown that processed meats, in particular, are viewed by the consumer to be an unhealthy product.

Both positive and negative nutritional attributes are associated with processed meat. Positive nutritional attributes include; a rich source of biologically valuable protein; contains an array of both vitamins and minerals; naturally low in both fat and sodium; contains multiple endogenous antioxidants and other bioactive compounds. (Williams, 2007; Jimenez-Colmenero, Carballo & Cofrades, 2001). Negative nutritional attributes associated with meat and meat products include; high levels of saturated fat; cholesterol; high levels of sodium and other harmful chemicals (Chapter 3; Whitney & Rolfes, 2002). However, many of these concerns are being addressed by the meat industry and regulatory authorities. Consequently, a great deal of research has been conducted (Tobin, O'Sullivan, Hamill, & Kerry, 2012a, 2012b, 2013; Yang, Choi, Jeon, Park, & Joo, 2007; Solheim & Ellekjær, 1993) to reduce or replace fat in processed meats. Sodium reduction has also been studied (Tobin et al, 2012a, 2012b,

2013; Ruusunen, Vainionpaa, Lyly, Lahteenmaki, Niemisto & Ahvenainen, 2005; Maurer, 1983). Further research investigating nitrite reduction in processed meats has also been investigated (Viuda-Martos, Fernández-López, Sayas-Barbera, Sendra, Navarro, & Pérez-Álvarez, 2009; Dineen, Kerry, Lynch, Buckley, Morrissey, & Arendt, 2000)

By reformulating meat products to enhance their health status the meat industry is hoping to change consumer perception towards processed meats. Additionally, the meat industry is starting to develop functional meat products. The functional food industry is one of the most rapidly growing sectors within the food industry. It has been estimated that by 2015 it shall be worth \$130 billion globally (Global Industry Analysts, 2010). Functional foods have previously been defined by Jimenez-Colmenero et al. 2001, as a product that is derived from naturally-occurring ingredients; consumed daily as part of an overall diet and provides health benefits beyond basic nutrition.

Meat-based functional foods can be created using a range of strategies from supplementation of animal feed to addition of a functional ingredient to an existing product. Goldberg (1994) identified 12 broad groups of ingredients which can be considered as bioactive or functional and many of these are readily available in market to be used to create new products. Co-enzyme Q10 (CoQ10) is just one example of a functional bioactive that could be incorporated into processed meat products.

CoQ10 has garnered great interest amongst the scientific community due to the potential health benefits it can offer and its importance in the human body (Rosenfeldt, Marasco, Lyon, Wowk, Sheeran, Bailey, Esmore, Davis, Pick, Rabinov, Smith, Nagley & Pepe, 2005; Chew & Watts, 2004; Rosenfeldt, Pepe, Linnane,

Nagley, Rowland, Ou, Marasco & Lyon, 2002; Lockwood, Moesgaard & Folkers, 1994). CoQ10 is naturally found in meat, poultry and fish (Stocker, 2007). However, the level of CoQ10 in these foods is too low to achieve any beneficial effect in the body (Weber, Bysted & Holmer, 1996).

The objective of this study is to compare the consumer sensory evaluation of reduced salt and fat processed meat products against commercially available products, and also to investigate consumer sensory acceptance of meat products enriched with CoQ10.

9.2 Materials and Methods

9.2.1 Sample Preparation

Beef and pork were selected on the basis of a high visual lean (V/L) score, (FSA, 2003); pork shoulder was used with a V/L score of 99% and beef shin was used with a V/L score of 95%. Beef and pork were purchased along with pork back fat from a local supplier (Ballyburden Meats Ltd., Ballincollig, Cork, Ireland). The meat and fat were vacuum-packed and stored at -18°C until required for product production.

For sausage production, frozen meat and fat was cut into strips and allowed to thaw slightly before being minced through a 5 mm plate (TALSABELL S. A., Spain). The reduced salt and fat variant (RS) was determined from previous work by Tobin et al. (2013) who's research work found this formulation (22.5% Fat, 1.4% Salt) to be consumer optimal from experimental products ranging in fat (22.5% to 37.5%) and salt levels (0.8% to 2.4%). The meat was weighed according to the formulations shown in Table 1 and fed into a bowl chopper (Maschinenfabrik, Seydelmann, Stuttgart, D70174). NovaSolQ[®] (AquaNova[®] AG, Darmstadt, Germany) was used as

the source of CoQ10 for both products. For the CoQ10 enriched samples the required NovaSolQ[®] was added to the pork in the bowl chopper also. The required salt, seasoning and half the water was added and mixed at high speed for 60 seconds. The required fat was then added and the mix was chopped for further 60 seconds at high speed. The remaining water and rusk was then added and mixed at low speed for 15 seconds and high speed for 30 seconds. The sausage mix was then put into the casing filler and fed into collagen casings (DEVRO, Scotland Ltd Moodiesburn, Glasgow, G690JE). The sausages were then sealed into laminated plastic bags of Polyamide/Polyethylene and stored in chill over-night at -4°C.

For Frankfurter manufacture, the frozen meat and fat were cut into strips and allowed to thaw slightly before being minced through a 3 mm plate (TALSABELL S. A., Spain). The reduced fat variant (RS) was determined from precious work by Tobin et al. (2013) who's research work found this formulation (10% Fat, 2.4% Salt) to be consumer optimal from experimental products ranging in fat (10 % to 25 %) and salt levels (1 % to 2.5 %). The meat was weighed according to the formulations shown in Table 1 and fed into the bowl chopper. The required NovaSolQ[®] was then added to the meat in the bowl chopper for the CoQ10 enriched samples. The required salt, nitrite and two thirds of the water was added and mixed at high speed (3000 r.p.m.) for 60 seconds. The required fat and seasoning was then added and the mix was chopped for further 120 seconds at high speed. The remaining water was then added and mixed at high speed for 45 seconds. The Frankfurter mix was then placed into the casing filler and stuffed into cellulose casings (Teepak Wienie-pak, Wysogotowok Poznania, ul Bukowska 18, 62-081 Przeźmierowo). The Frankfurters were hung in a Zanussi convection oven (C. Batassi, Conegliano, Italy) and cooked at 90°C until an internal temperature of 72°C tested by a temperature probe (Testo 110, Lenzkirch, Germany) was reached, the Frankfurters were then held at 72°C for 10 minutes. All test samples were cooked at the same time and segregated to prevent any mixing. The Frankfurters were then sealed into polyamide/polyethylene (PA/PE) laminate plastic bags and stored in the chill overnight at -4°C.

9.2.2 NovaSol $Q^{\mathbb{B}}$

NovaSolQ[®] was used in order to create the CoQ10 fortified products. NovaSolQ[®] has already achieved a GRAS status and also supplies the CoQ10 in a clear solution and using an amphiphilic micelle structure helps to protect the CoQ10 from environmental stresses. The micelles diameter was given at less than 30 nonometers and reported by the manufacturer to require no further biological micellization to pass through the small intestine.

9.2.3 Commercial Products

Two leading commercially available products for sausages and Frankfurters were purchased from supermarkets found throughout Ireland. Compositional analysis was carried out on each product in triplicate; the composition of these products is shown in Table 1.

9.2.4 Cooking

Oven-cooking was used to cook all samples as it provided greater control and was found to be the most repeatable cooking method compared to other options. The sausages (including commercial brands) were wrapped in foil and dry-cooked at 150°C in a Zanussi convection oven (C. Batassi, Conegliano, Italy) for approximately 15 min to an internal temperature of 73°C, as measured by an internal temperature probe (Testo 110, Lenzkirch, Germany). All test samples were cooked at the same time in insure uniformity and segregated to prevent mixing. Frankfurters were all precooked and required only preheating before sensory evaluation.

9.2.5 Sensory Evaluation

Sensory analysis on the products was carried out using 100 regular consumers of sausage style meat products. Panellist's ages ranged within 18-65 years old. All analysis was undertaken in the university sensory laboratory, using panel booths that conform to ISO (1988) international standard. Panellists were presented with eight samples in total. Four samples were presented to the panellist on a plate to begin, after a short break the further four samples were presented. Sample presentation order was also randomised in order to prevent any flavour carryover effects (MacFie, Bratchell, Greenhoff & Vallis, 1989). Participants were required to rinse their mouths with water before tasting each sample. Consumers were asked to asked their rate on a 10 cm line scale ranging from 0 at the left to 10 at the right and rating subsequently scored in cm from left for each sausage and Frankfurter sample presented (O'Sullivan, Byrne, and Martens, 2003). Using the following descriptors consumers were requested to evaluate every sample: appearance, texture, tenderness, juiciness, salt taste, meat flavour, fat flavour, overall flavour intensity, off-flavour, oxidative flavour and overall acceptability. Oxidative flavour was described to consumers as rancid, cardboard or linseed oil-like flavour.

9.2.6 Protein Content

The Kjeldahl method (Suhre, Corrao, Glover & Malanoski, 1982) was used to measure protein concentrations. A digestion block was pre-heated to 410°C. Samples were well homogenised and approximately 0.5 g was weighed accurately into a digestion tube. 15ml of sulphuric acid (nitrogen free), 10ml hydrogen peroxide and 2 "kjeltabs" were added to the sample. The tubes where then placed into the heated digestion block. When the samples became colourless they were removed from the block. The tubes were allowed to cool in the fume hood after removal.

50 ml of distilled water was carefully added to the cooled and digested sample inside the fume-hood. The tubes and a receiver flask containing 50 ml of 4% Boric acid with indicator were then placed into the distillation unit. After the sample had been distilled the contents of the receiver flask were titrated against 0.1 N hydrochloric acid until the green colour reverted back to the original red colour.

9.2.7 Ash Content

Ash content was determined by muffle furnace (AOAC, 1923). A muffle furnace was pre-heated to 525°C. Approximately 5.000 g of well homogenised sample were weighed into porcelain dishes using a balance that weighs to 1 mg. The sample containing dishes were then put in the muffle furnace for (approximately 6 hours) until the colour of the samples went white. The dishes were then removed and placed in desiccators to cool. The new weight of dishes was then noted and the ash content calculated.

9.2.8 Moisture and Fat Content

A Büchi Mixer B-400 (BÜCHI Labortechnik AG, Meierseggstrasse 40, Postfach, CH-9230 Flawil 1, Switzerland) was used to homogenise a total of 200g of sample. To avoid moisture or evaporative loss the homogenised sample was then quickly transferred into a moisture proof bag Moisture and fat content were then determined using the CEM SMART (moisture) and SMART Trac (fat) systems (Bostian, Fish, Webb, and Arey, 1985).

9.2.9 Colour

Both raw and cooked samples for sausages were measured for colour while only cooked Frankfurter sample were measure for colour due to pre-cooking. Measurements for colour were carried out according to the CIE L* a* b* colour system, both sausages and Frankfurters were cut down the centre and the interior colour measured. Cooked samples were cooled to room temperature before measuring. A Minolta CR 300 colorimeter (Minolta Camera Co. Ltd., Osaka, Japan) with an 11 mm-diameter aperture, D₆₅ illuminant, calibrated by the CIE Lab colour space system using a white tile (C: Y = 93.6, x = 0.3130, y = 0.3193), (Minolta calibration plate) was used to conduct analysis. Colour measurements (CIE L^* , a^* and b^* values representing lightness, redness and yellowness, respectively) were taken. Nine readings were taken per sample.

9.2.10 Cooking Loss

Sausage sample weights were recorded before and after cooking and the differences in weights recorded. The samples were cooked as described in 2.2. Before weighing, samples were blotted with a paper towel to remove excess surface moisture.

Calculation for cook loss was as follows:

% cook loss = ((cooked weight – raw weight)/raw weight) x 100

9.2.11 Texture Analysis

Texture measurements were obtained for individual samples using texture profile analysis (TPA). All analysis was performed at room temperature with a Texture Analyser 16 TA-XT2i (Stable Micro Systems, Surrey, UK). Sausage and Frankfurter were cut into (10 mm x 10 mm) cylindrical slices prior to analysis. Every slice was then subjected to a two-cycle compression test using the 25 kg load cell. Samples were compressed to 40% of their original height with a cylindrical probe (SMSP/35 Compression plate) 35 mm diameter and a cross-head speed of 1.5 mm/sec. All analyses for sausage and Frankfurter samples were performed in triplicate.

Descriptions by Bourne (1978) and the SMS manual (Stable Micro Systems, Surrey, UK), were used to determine texture profile parameters, these included; hardness (N) maximum force required for the initial compression of the sample; springiness (mm), the samples ability to recover its original shape after the initial compression and the deforming force was removed; adhesiveness ($N \times mm$), area under the abscissa post initial compression; cohesiveness (Dimensionless), extent to which the sample could be deformed prior to rupture, measured by the areas under the compression portion only and excludes the areas under the decompression portion instead of using the total area under positive force; chewiness ($N \times mm$), the required work to masticate the sample, measured as the product of hardness times cohesiveness times springiness; and resilience (*Dimensionless*), the ratio between the negative force input to positive force input during the first compression.

9.2.12 Salt Determination

Salt levels were determined using the Volhard method for salt determination (AOAC, 1995) in each product. Approximately 2.5 to 3 g of sample was weighed into each conical flask. 15 ml of concentrated Nitric acid and 25 ml of 0.1 N Silver Nitrite solutions were added. Sufficient amounts of boiling chips were included to the flasks and the samples were boiled until the meat digested. Potassium Permanganate was added in small doses while boiling until the solutions maintained the dark colour for several minutes before reverting back to clear. Next, 25 ml of distilled water was added to the flasks and the solutions continued to boil for approximately 5 min before being removed from the heat source and allowed to cool to room temperature. The samples were then diluted to approximately 150 ml with distilled water. 2 ml of ferric alum indicator was then added to each sample and excess Silver Nitrite was titrated against Potassium Thiocyanate.

9.2.13 Data Analysis

All data accumulated from the 100 panellists during sensory evaluation and data acquired from instrumental methods were processed using ANOVA-partial least squares regression (APLSR). APLSR, is based on cross-validation and stability plots (Martens & Martens, 1999, 2001). All analyses were performed using the Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway). The X-matrix was designed as 0/1 design variables for the four sausages in Figure 1 and Frankfurters in Figure 2. Sensory, chemical, and instrumental variables were used for the Y-matrix design.

Four principal components were found to be the optimal number of components in the APLSR models presented for both figures; PC 1 versus PC 2 is also presented for both of the Figures; the other PCs did not yield any other additional information or provide any predictive improvement in the Y-matrix obtained prior examination. The validated explained variance for the model constructed was 26.63 % and the calibrated variance was 26.92 % for Figure 1; and 18.84% validated explained variance and 19.16% calibrated variance for Figure 2.

The significant effects of consumer sensory evaluation were carried out on the raw sensory data and analyzed using the General Linear Model (GLM) with post hoc Duncan comparison of mean scores (SPSS, Chicago, IL, USA). A (P<0.05) significance level was set for all analysis. These values are presented in Table 2 for sensory variables and Table 3 for physiochemical variables.

9.3 Results and Discussion

9.3.1 Compositional Analysis

The results from the Consumer sensory evaluation (n=100) and instrumental analysis are presented in the APLSR plots in Figure 1 for sausages, and Figure 2 for Frankfurters. The mean values for sensory variables are shown in Table 2, and physiochemical data is shown in Table 3.

Formulations for the non-commercial sausages and Frankfurters were determined from previous work conducted by Tobin et al. (2012b) and (2013) respectively. Consumer sensory analysis carried out by Tobin et al. (2013) found that sausages containing lower fat and salt were significantly preferred by consumers. Tobin et al. (2012b) investigated Frankfurter formulations and found that consumers tended to prefer higher salt but lower fat Frankfurters.

The Compositional analysis of each sample was carried out after cooking and is displayed in Table 1. Cooking loss is primarily caused by the cook-out of fat and moisture in meat products. From Table 1 it can be observed that reduced salt and fat products and the CoQ10 fortified products had less (P < 0.05) fat when compared to commercial products (Table 1) and higher moisture levels (Table 1). Cook-loss was not measured for Frankfurters as these products are sold pre-cooked (Table 3).

Cooking-loss was higher (P<0.05) for commercial sausage 1 (CS1) and commercial sausage 2 (CS2) compared to the reduced salt and fat sausage (RS) and the CoQ10-enrinched sausage (CoS) (Table 3). This was likely caused by the reduction of fat present in these samples. Previous research carried out by Carballo, Mota, Barreto & Jimenez-Colmenero (1995) showed that sausages containing more protein and less fat had less cooking loss. Banon, Diaz, Nieto, Castillo & Alvarez (2008) indicated that the increase in fat to lean ratio in comminuted pork products caused a significant increase in cooking-loss compared to products with lower levels of fat. Previous research has shown that salt levels play a significant role in cooking loss. Tobin et al. (2012a, 2013) and Ruusunen et al. (2005) found that increasing salt levels in meat products reduced cooking loss. Salt, is used by industry to solubilise myofibrillar protein and consequently to assist in the binding of water and in the encapsulation of fat within the matrix of the meat products This statement is supported from the significant (P<0.05) difference observed between cooking-loss values for CS1 and CS2.

The reduced salt and fat products, as well as those containing CoQ10 fortified products, were found to contain higher (P<0.05) levels of moisture and protein (Table 1). The higher protein concentration and water content in experimentally produced sausages (RS, CoS) was as a result of higher levels of lean meat being employed in the manufacture of the reduced fat products. Lean meat is known to contain more water than fattier meat or that containing a higher connective tissue content, this, coupled with a higher myofibrillar content in lean meat, provides for a stronger water encapsulating gel network binding more water in the product following salt activation of the myofibrillar proteins (Foegeding & Lanier, 1987; Acton & Dick, 1984).

9.3.2 Sensory and Instrumental Evaluation of Sausages

Figure 1 displays an ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for sausage sensory and instrumental data. Table 2 displays the mean values for sensory data with significances determined from Duncan GLM. CS1 scored higher (P<0.05) in terms of appearance compared to CS2 (Table 2). No significant difference was determined between the other sausage treatments in terms of appearance. CIE L* a* b* colour measurements for CS2 (Table 3) were highest (P<0.05) for a* value compared to all other samples; the greater a* value likely to be

due to the increased levels of salt found within the sample. Ventanas, Puolanne, & Tuorila (2010) in a recent study on bologna type sausages found that consumers perceived more intense colour in higher salt sausages. Significant (P<0.05) differences were also found between b* values for RS and CoS treatments, with CoS presenting a more (P<0.05) yellow hue than other sausage samples. However, no negative impact on consumer acceptance of appearance was found when compared to instrumental analysis. This concurs with data generated by this Chapter 8 in relation to similar work conducted for beef patties who reported an increase in b* values in beef patties fortified with NovaSolQ[®].

CS2 was less (P < 0.05) tender than all other sausage treatments. RS also had a lower tenderness score which is evident in Figure 1, however, it was not found to be less (P < 0.05) acceptable than CoS and CS1 (Table 2). The most tender products were CS1 and CoS, CS1 was found to be significantly (P<0.05) more tender than RS and CS2, but not more so than CoS (Table 2). This is likely due to the higher level of fat found within CS1 and the lubricating effect of fat within the meat system (Javidipour, Vural, Ozbas & Tekin, 2005; Crehan, Troy & Buckley, 2000; Giese, 1996).

CS1 was the highest rated product for juiciness (Figure 1), and was also found to be more (P<0.05) juicy than RS and CoS. Fats are known to assist in texture development, mouth feel properties and assist in the overall sensation of lubricity in foods (Giese, 1996). The higher level of fat in CS1 explains the reason why consumers found CS1 to be juicier than the other samples. No significant difference were found in juiciness between CoS and RS.

The only statistical differences determined for salt perception was between the highest scoring product CS1 and the lowest CS2. RS and CoS treatments were not

significantly different from either of the commercial products (Table 2). Previous studies have shown that higher fat content in meats tend to increase sensory salt perception (Tobin et al. 2013; Ruusunen, Simolin & Puolanne, 2001; Tuorila, Cardello & Lesher, 1994). Table 2 shows that CS1 scored higher (P < 0.05) for meat flavour than the other three sausage products. Perhaps this is due to the higher salt and fat levels present in this product compared to RS and CoS, both known to be flavour enhancing ingredients (Durack et al. 2008; Silva et al. 2003). In Figure 1, fat flavour was more highly correlated to CS1 and CS2 than to RS and CoS. These products were also shown to have the highest fat levels (Table 1). Table 2 shows a higher (P<0.05) scoring in this attribute for CS2 compared to RS and CoS. No significant differences were found for overall flavour intensity between RS, CoS and CS2, however, CS1 (Table 2) scored higher (P<0.05) for overall flavour intensity by panellists. The differences in flavour perception for all sausage products can be attributed to gross compositional differences pertaining to protein, fat and moisture content, as well as fine differences relating to individual product seasoning differences. The higher scores for CS1 can also be attributed to the higher fat and salt levels, both of which function as flavour enhancers in meat products (Tobin et al. 2012b; Silva, Morais & Silvestre, 2003)

None of the products scored differently (P<0.05) for off-flavour or oxidative flavour. CS1 scored higher (P<0.05) for Overall acceptability compared to the other sausages (Table 2), the next highest being CoS and RS, however, there were no significant differences between these products and CS2. No significant differences were found between CoS and RS for flavour intensity or overall acceptability.

9.3.3 Sensory and Instrumental Data for Frankfurters

Figure 2 displays an ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for Frankfurter sensory and instrumental data. Table 2 displays mean values for sensory data with significances determined from Duncan GLM. Salt levels for Frankfurters were not reduced in this study due to previous work undertaken by Tobin et al. (2012b) who found that lowering the salt content had unfavourable effects on Frankfurter quality, particularly in relation to texture. However, the formulation used was the optimal from the ranges studied from a consumer perspective. The panellists scored the appearance of commercial Frankfurter 1 (CF1) and commercial Frankfurter 2 (CF2) significantly (P<0.05) higher than for Reduced fat Frankfurters (RF) and the CoQ10 enriched Frankfurter (CoF). This is also clearly evident in the bottom right quadrant of Figure 2. The L* value and a* value of the RF and CoF products were found to be higher (P<0.05) than for commercial products (Table 3); the more pronounced red colour and darker appearance likely to be due to the higher lean meat content of these Frankfurters, as lean meat is more red in colour and darker in appearance than meat cuts which contain much higher levels of fat (Tobin et al, 2012b). As with sausages (described above) CoF products had higher (P<0.05) b* values than observed for the remaining three Frankfurter variants. This was most likely due to the yellow hue associated with the ingredient co-enzyme Q 10, the yellow colour however was not shown to have any negative impact on consumer preference.

Liking of texture is shown in Figure 2 to be highly correlated to CF1 and CF2, Table 2 shows that CF1 and CF2 scored higher (P<0.05) than RF and CoF. RF received the highest score for tenderness, followed by CF2, both of which scored higher (P<0.05) for tenderness compared to CoF (Table 2), but not significantly higher when compared with CF1. Previous research has also shown that lower fat Frankfurters were less tender than higher fat products (Cengiz & Gokoglu, 2007; Mittal & Barbut, 1994; Hand, Hollingswort, Calkins, & Mandigo, 1987). Juiciness (Figure 3) was found to be highly correlated to CF1 and CF2; (Table 2); both scoring higher (P<0.05) for juiciness than RF and CoF products. A decrease in juiciness scores due to reduction in fat levels has previously been reported (Tobin et al. 2012b; Mittal & Barbut, 1994; Berry & Leddy, 1984) for low fat beef patties and Frankfurters.

None of the products were found to be significantly different in terms of saltiness, as the Frankfurters used in this study were not reduced in salt. CF2 scored significantly higher for meat flavour compared to RF and CoF, but not when compared to CF1. CF1 scored higher (P<0.05) than CoF, which scored lowest in terms of meat flavour. The higher fat content of CF1 and CF2 is the most likely reason for the increased perceived meat flavour by consumers. Interactions between fat and other ingredients are known to enhance and develop overall flavour (Javidipour et al. 2005; Crehan et al. 2000; Giese, 1996). None of the products were significantly different for fat flavour and oxidative flavour when compared to the other products.

RF samples scored the highest for off-flavour and scored differently (P<0.05) when compared to CF2 by panellists. CF2 was rated highest in terms of overall acceptability, but was not rated higher than CF1. However, differences (P<0.05) were present between the two commercial Frankfurters and the two experimental treatments. Very few differences were found between CoF and RF in consumer sensory evaluation. CoF was less (P < 0.05) tender than RF, however, both variants were similarly scored for juiciness and texture (Table 2). Although RF and CoF

treatments were found to be less acceptable to the consumer panel than commercially available products, overall acceptability between these samples was not significantly different. This indicates that CoQ10 could be successfully added to an existing commercial brand of Frankfurter, thereby increasing its nutritional benefit, while having minimal adverse affects on quality. The improved sensory performance of the commercial products, particularly for overall acceptability was also due to the incorporation of a more commercially recognisable flavouring formulation used compared to the generic and simple spice mix used in RF and CoF products.

9.4 Conclusion

In conclusion, by reducing fat and salt in sausages, it is possible to produce products which consumers find to be as acceptable as commercially available products. Reduced salt/fat sausages fortified with CoQ10 can be produced which do not negatively affect the acceptance level of the consumer. Frankfurters with reduced fat levels were not found to be acceptable to consumers, however, CoQ10 maybe successfully added to an existing commercial brand of Frankfurter without deleteriously affecting the flavour profile, thus changing that product into a functional food.

9.5 Acknowledgements

This work was funded by the Irish Food Industry Research Measure (FIRM) as part of the project titled 'Understanding the physiochemistry of the meat matrix and its potential as a delivery system for added bioactive compounds'.

9.6. Tables and Figures.

Table 1. Formulations and Compositional analysis.

		Sample Formulation							
Sample	% Beef	% Pork Fat	% Pork	% Salt	% Water	% Rusk	% Seasoning	% Nitrite	NovaSolQ %
Reduced Fat and Salt Sausages	0	22.5	42.15	1.4	20	12.5	1	0	0
CoQ10 Enriched Sausages	0	22.5	42.15	1.4	20	12.5	1	0	0.45
Reduced Fat Frankfurters	35	10	35	2.5	20	0	0.75	0.02	0
CoQ10 Enriched Frankfurters	35	10	35	2.5	20	0	0.75	0.02	0.45

			Comp	ositional Analysis		
	Code	Moisture	Fat	Protein	Carbohydrate	Salt
Sausage 22.5%Fat 1.4%Salt	RS	56.76 ± 0.18^{a}	14.03 ± 0.17^{a}	14.27 ± 0.31^{a}	13.53 ± 0.01^{a}	1.40 ± 0.01^{a}
Sausage CoQ10	CoS	50.85 ± 0.11^{b}	$18.38 \ \pm \ 0.09^{b}$	14.38 ± 0.64^{a}	14.24 ± 0.01^{a}	1.42 ± 0.01^{a}
Commercial Sausage 1	CS1	$45.45 \pm 0.21^{\circ}$	28.38 ± 0.22^{c}	11.56 ± 0.16^{c}	12.96 ± 0.01^{a}	1.65 ± 0.01^{b}
Commercial Sausage 2	CS2	$33.01 ~\pm~ 1.43^{d}$	$26.57 ~\pm~ 1.09^{d}$	$10.43 \pm 0.49^{\circ}$	$21.34~\pm~0.01^{a}$	$2.00~\pm~0.01^{c}$
Frankfurter 10%Fat 2.5%Salt	RF	62.43 ± 0.71^{a}	14.61 \pm 0.18 ^a	$21.01 ~\pm~ 0.35^{a}$	$0.01~\pm~0.01^{a}$	$2.50 ~\pm~ 0.01^{a}$
Frankfurter CoQ10	CoF	62.87 ± 0.33^{a}	14.00 ± 0.15^{b}	20.95 ± 0.16^{a}	$0.01 ~\pm~ 0.01^{a}$	$2.51 ~\pm~ 0.01^{a}$
Commercial Frankfurter 1	CF1	56.32 ± 0.01^{b}	26.35 ± 0.19 ^c	13.45 ± 0.17^{b}	1.05 ± 0.01 ^a	$2.83~\pm~0.01^{b}$
Commercial Frankfurter 2	CF2	52.58 ± 0.43 ^c	32.91 ± 0.17^{d}	11.83 \pm 0.35 ^c	1.11 ± 0.01 ^a	2.56 ± 0.01^{a}

	Appearance	Texture	Tenderness	Juiciness	Salt Taste	Meat Flavour	Fat Flavour	Overall Flavour	Off Flavour	Oxidative	Overall
	Appearance	Texture	Tondomode	Galomooo	Our ruoto	Moat havou	i at i avoai	Intensity	On Flavoa	Flavour	Acceptability
Sausage 22.5%Fat 1.4%Salt	$4.82~\pm~0.25^{ab}$	$6.27 ~\pm~ 0.23^a$	$6.70~\pm~0.20^{\text{ac}}$	6.16 ± 0.21^{ac}	6.14 ± 0.22^{abc}	$5.93~\pm~0.20^a$	5.98 ± 0.19^{a}	$6.06~\pm~0.24^a$	$3.90~\pm~0.24^a$	$3.80~\pm~0.22^a$	5.74 ± 0.24^{a}
Sausage CoQ10	$4.40~\pm~0.24^{ab}$	$5.97 \ \pm \ 0.24^{a}$	$7.32~\pm~0.19^{abc}$	$5.98 \ \pm \ 0.19^{a}$	$6.12~\pm~0.21^{abc}$	$5.67 ~\pm~ 0.21^{a}$	$5.69 \ \pm \ 0.21^{a}$	$6.08~\pm~0.23^a$	$3.91~\pm~0.25^a$	$3.67 \ \pm \ 0.21^{a}$	$5.93 \ \pm \ 0.22^{a}$
Commercial Sausage 1	$5.14 \ \pm \ 0.25^{a}$	$6.18~\pm~0.25^a$	7.88 ± 0.16^{b}	$7.30 \ \pm \ 0.19^{b}$	$6.92~\pm~0.22^b$	$6.69~\pm~0.18^{b}$	$6.34~\pm~0.21^{ab}$	$6.99~\pm~0.22^{b}$	$3.77 ~\pm~ 0.24^{a}$	$3.77 \ \pm \ 0.22^{a}$	$6.76 \ \pm \ 0.24^{b}$
Commercial Sausage 2	$4.16~\pm~0.25^{\texttt{b}}$	$5.06 \pm 0.25^{\text{b}}$	$6.85~\pm~0.20^{c}$	$6.87 ~\pm~ 0.20^{\text{bc}}$	$5.49 ~\pm~ 0.24^{c}$	$5.74 ~\pm~ 0.23^a$	$6.84 \pm 0.23^{\texttt{b}}$	$5.64~\pm~0.23^a$	$3.86~\pm~0.24^a$	$3.68~\pm~0.22^a$	$5.32~\pm~0.20^{a}$
Frankfurter 10%Fat 2.5%Salt	4.30 ± 0.21^{a}	4.56 ± 0.24^{a}	7.67 ± 0.20^{a}	5.71 ± 0.23^{a}	6.80 ± 0.23^{a}	$5.97 ~\pm~ 0.23^{\text{ab}}$	5.45 ± 0.22^{a}	5.24 ± 0.25^{a}	4.45 ± 0.29^{a}	3.85 ± 0.23^{a}	4.88 ± 0.24^{a}
Frankfurter CoQ10	4.73 ± 0.21^{a}	$4.72 \ \pm \ 0.22^{a}$	$6.66~\pm~0.20^{b}$	$5.27 \ \pm \ 0.20^{a}$	$6.72 \ \pm \ 0.22^{a}$	$5.61~\pm~0.20^a$	5.10 ± 0.21^{a}	$4.65~\pm~0.22^a$	$4.25~\pm~0.28^{ab}$	$3.67\ \pm\ 0.22^a$	$4.58 \ \pm \ 0.21^{a}$
Commercial Frankfurter 1	$7.05~\pm~0.23^{\text{b}}$	$6.53 ~\pm~ 0.25^{\text{b}}$	$7.05~\pm~0.20^{ab}$	$7.06~\pm~0.20^{b}$	$7.15 \ \pm \ 0.22^{a}$	$6.51~\pm~0.21^{\text{bc}}$	$5.58 ~\pm~ 0.21^{a}$	$7.00~\pm~0.24^{b}$	$3.77~\pm~0.26^{ab}$	$3.54~\pm~0.23^a$	$6.93~\pm~0.23^{b}$
Commercial Frankfurter 2	$7.32~\pm~0.22^{\texttt{b}}$	$7.01 ~\pm~ 0.23^{\texttt{D}}$	7.43 ± 0.18^{a}	$6.86~\pm~0.18^{\texttt{b}}$	$6.35 \hspace{0.1in} \pm \hspace{0.1in} 0.21^{a}$	$6.92~\pm~0.18^{\rm c}$	$5.34 0.20^a$	$7.06~\pm~0.23^{\texttt{D}}$	$3.37 \hspace{.1in} \pm \hspace{.1in} 0.23^{\scriptscriptstyle D}$	$3.42~\pm~0.21^{a}$	$7.44 \pm 0.20^{\circ}$

Table 2. Sensory Mean Values and Standard Deviation.

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b, c, d means with different letters in columns are significantly different (p < 0.05). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.

Table 5. Filyslo-chemic	al Analysis M	ean values a	ing Standard	Deviation.				-
	Cooking Loss	Raw L value	Raw a value	Raw b value	Cooked L value	Cooked a value	Cooked b value	Cooked Salt
Reduced Salt/Fat Sausage	0.05 ± 0.01^{a}	75.80 ± 1.21 ^a	8.39 ± 0.68^{a}	16.99 ± 0.73^{a}	72.31 ± 1.40 ^a	3.25 ± 0.38^{a}	17.07 ± 0.65^{a}	1.40 ± 0.01^{a}
Sausage CoQ10	0.09 ± 0.01^{b}	68.23 ± 1.39 ^b	7.21 ± 0.35^{b}	27.98 ± 0.77 ^b	65.32 ± 0.84^{b}	3.07 ± 0.23^{b}	22.17 ± 0.40^{b}	1.42 ± 0.01 ^a
Commercial Sausage 1	$0.16 + 0.02^{c}$	70.69 + 1.01 ^c	11.13 + 0.04 ^c	14.10 + 0.41 ^c	$68.06 + 0.70^{\circ}$	$3.41 + 0.06^{a}$	14.36 + 0.52 ^c	$1.65 + 0.01^{b}$
Commercial Sausage 2	0.12 ± 0.01^{d}	72.58 ± 1.71 ^d	7.38 ± 1.11 ^b	14.52 ± 0.22^{d}	66.88 ± 1.50^{d}	$7.89 \pm 0.89^{\circ}$	12.13 ± 1.18 ^d	$2.00 \pm 0.01^{\circ}$
Poducod Salt/Eat Frankfurtor		/		/	67.75 ± 1.32^{a}	12 22 ⊥ 0 32 ^a	11.67 ⊥ 0.36 ^a	250 ± 0.01^{a}
Fronkfurter CoO10					62.40 ± 3.00^{b}	10.03 ± 0.02^{b}	17.07 ± 0.30	2.50 ± 0.01
					62.40 ± 3.09	10.24 ± 0.27	17.00 ± 0.41	2.51 ± 0.01
Commercial Frankfurter 1					74.66 ± 0.47	9.13 ± 0.08	12.74 ± 0.11	2.83 ± 0.01
Commercial Frankfurter 2					71.48 ± 0.72	9.82 ± 0.18	13.05 ± 0.41	2.56 ± 0.01
	Cooked Moisture	Cooked Fat	Cooked Protein	Hardness	Springiness	Cohesiveness	Chewiness	Resilience
Reduced Salt/Fat Sausage	Cooked Moisture 56.76 \pm 0.18 ^a	Cooked Fat 14.03 ± 0.17 ^a	Cooked Protein 14.27 ± 0.31^{a}	Hardness 5346.27 ± 499.91 ^a	Springiness 0.93 ± 0.02^{a}	Cohesiveness 0.56 ± 0.01^{a}	Chewiness 2792.53 ± 385.95 ^a	Resilience 0.41 ± 0.02 ^a
Reduced Salt/Fat Sausage Sausage CoQ10	Cooked Moisture 56.76 ± 0.18^{a} 50.85 ± 0.11^{b}	Cooked Fat 14.03 ± 0.17 ^a 18.38 ± 0.09 ^b	Cooked Protein 14.27 ± 0.31^{a} 14.38 ± 0.64^{a}	Hardness 5346.27 ± 499.91 ^a 5004.34 ± 312.41 ^b	$\begin{array}{r} \text{Springiness} \\ 0.93 \pm 0.02^{a} \\ 0.87 \pm 0.03^{b} \end{array}$	$\begin{array}{l} \mbox{Cohesiveness} \\ \mbox{0.56} \pm \ 0.01^{a} \\ \mbox{0.54} \pm \ 0.01^{b} \end{array}$	Chewiness 2792.53 \pm 385.95 ^a 2325.82 \pm 229.15 ^b	Resilience 0.41 ± 0.02^{a} 0.30 ± 0.01^{b}
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1	Cooked Moisture 56.76 ± 0.18^{a} 50.85 ± 0.11^{b} 45.45 ± 0.21^{c}	Cooked Fat 14.03 ± 0.17^{a} 18.38 ± 0.09^{b} 28.38 ± 0.22^{c}	Cooked Protein 14.27 ± 0.31^{a} 14.38 ± 0.64^{a} 11.56 ± 0.16^{c}	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c}	$\begin{array}{r} \text{Springiness} \\ 0.93 \pm \ 0.02^{a} \\ 0.87 \pm \ 0.03^{b} \\ 0.86 \pm \ 0.03^{b} \end{array}$	$\begin{array}{r} \text{Cohesiveness} \\ 0.56 \pm \ 0.01^a \\ 0.54 \pm \ 0.01^b \\ 0.56 \pm \ 0.01^c \end{array}$	$\begin{array}{r} \mbox{Chewiness} \\ 2792.53 \pm \ 385.95^a \\ 2325.82 \pm \ 229.15^b \\ 1238.74 \pm \ 167.99^c \end{array}$	$\begin{array}{r} \text{Resilience} \\ 0.41 \ \pm \ 0.02^{a} \\ 0.30 \ \pm \ 0.01^{b} \\ 0.35 \ \pm \ 0.01^{c} \end{array}$
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1 Commercial Sausage 2	$\begin{array}{rrrr} \text{Cooked Moisture} \\ 56.76 \pm \ 0.18^{a} \\ 50.85 \pm \ 0.11^{b} \\ 45.45 \pm \ 0.21^{c} \\ 33.01 \pm \ 1.43^{d} \end{array}$	$\begin{array}{r} \mbox{Cooked Fat} \\ 14.03 \pm \ 0.17^a \\ 18.38 \pm \ 0.09^b \\ 28.38 \pm \ 0.22^c \\ 26.57 \pm \ 1.09^d \end{array}$	$\begin{array}{rrr} \mbox{Cooked Protein} \\ 14.27 ~\pm~ 0.31^a \\ 14.38 ~\pm~ 0.64^a \\ 11.56 ~\pm~ 0.16^c \\ 10.43 ~\pm~ 0.49^c \end{array}$	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c} 5321.19 ± 441.57^{a}	$\begin{array}{r} \text{Springiness} \\ 0.93 \pm 0.02^{a} \\ 0.87 \pm 0.03^{b} \\ 0.86 \pm 0.03^{b} \\ 0.89 \pm 0.04^{c} \end{array}$	$\begin{array}{rl} \text{Cohesiveness} \\ 0.56 \pm \ 0.01^a \\ 0.54 \pm \ 0.01^b \\ 0.56 \pm \ 0.01^c \\ 0.58 \pm \ 0.01^d \end{array}$	$\begin{array}{r} \mbox{Chewiness} \\ 2792.53 \pm 385.95^a \\ 2325.82 \pm 229.15^b \\ 1238.74 \pm 167.99^c \\ 2776.96 \pm 264.46^a \end{array}$	$\begin{array}{r} \text{Resilience} \\ 0.41 \ \pm \ 0.02^{a} \\ 0.30 \ \pm \ 0.01^{b} \\ 0.35 \ \pm \ 0.01^{c} \\ 0.41 \ \pm \ 0.01^{a} \end{array}$
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1 Commercial Sausage 2	Cooked Moisture 56.76 ± 0.18^{a} 50.85 ± 0.11^{b} 45.45 ± 0.21^{c} 33.01 ± 1.43^{d}	Cooked Fat 14.03 ± 0.17^{a} 18.38 ± 0.09^{b} 28.38 ± 0.22^{c} 26.57 ± 1.09^{d} 14.61 ± 0.18^{a}	Cooked Protein 14.27 ± 0.31^{a} 14.38 ± 0.64^{a} 11.56 ± 0.16^{c} 10.43 ± 0.49^{c} 21.01 $\pm 0.35^{a}$	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c} 5321.19 ± 441.57^{a} 2257.62 ± 61.70^{a}	$\begin{array}{r} \text{Springiness} \\ 0.93 \pm \ 0.02^{a} \\ 0.87 \pm \ 0.03^{b} \\ 0.86 \pm \ 0.03^{b} \\ 0.89 \pm \ 0.04^{c} \end{array}$	Cohesiveness 0.56 ± 0.01^{a} 0.54 ± 0.01^{b} 0.56 ± 0.01^{c} 0.58 ± 0.01^{d}	Chewiness 2792.53 ± 385.95^{a} 2325.82 ± 229.15^{b} 1238.74 ± 167.99^{c} 2776.96 ± 264.46^{a} 1212.79 ± 63.33^{a}	Resilience 0.41 ± 0.02^{a} 0.30 ± 0.01^{b} 0.35 ± 0.01^{c} 0.41 ± 0.01^{a}
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1 Commercial Sausage 2 Reduced Salt/Fat Frankfurter	Cooked Moisture 56.76 ± 0.18^{a} 50.85 ± 0.11^{b} 45.45 ± 0.21^{c} 33.01 ± 1.43^{d} 62.43 ± 0.71^{a} $c_{2}0.27 \pm 0.22^{b}$	Cooked Fat 14.03 ± 0.17^{a} 18.38 ± 0.09^{b} 28.38 ± 0.22^{c} 26.57 ± 1.09^{d} 14.61 ± 0.18^{a} 14.00 ± 0.47^{cb}	Cooked Protein 14.27 ± 0.31^{a} 14.38 ± 0.64^{a} 11.56 ± 0.16^{c} 10.43 ± 0.49^{c} 21.01 ± 0.35^{a} 20.05 ± 0.46^{a}	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c} 5321.19 ± 441.57^{a} 2357.63 ± 61.70^{a} 2049.00 ± 50.00^{b}	$\begin{array}{r} \text{Springiness} \\ 0.93 \pm 0.02^{a} \\ 0.87 \pm 0.03^{b} \\ 0.86 \pm 0.03^{b} \\ 0.89 \pm 0.04^{c} \\ \end{array}$	$\begin{array}{r} \text{Cohesiveness} \\ 0.56 \pm \ 0.01^{a} \\ 0.54 \pm \ 0.01^{b} \\ 0.56 \pm \ 0.01^{c} \\ 0.58 \pm \ 0.01^{d} \\ \end{array}$	$\begin{array}{r} \mbox{Chewiness} \\ 2792.53 \pm 385.95^a \\ 2325.82 \pm 229.15^b \\ 1238.74 \pm 167.99^c \\ 2776.96 \pm 264.46^a \\ 1312.78 \pm 63.33^a \\ 1302.45 \pm 69.45^b \end{array}$	Resilience 0.41 ± 0.02^{a} 0.30 ± 0.01^{b} 0.35 ± 0.01^{c} 0.41 ± 0.01^{a} 0.20 ± 0.01^{a}
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1 Commercial Sausage 2 Reduced Salt/Fat Frankfurter Frankfurter CoQ10	Cooked Moisture 56.76 ± 0.18^{a} 50.85 ± 0.11^{b} 45.45 ± 0.21^{c} 33.01 ± 1.43^{d} 62.43 ± 0.71^{a} 62.87 ± 0.33^{b}	Cooked Fat 14.03 ± 0.17^{a} 18.38 ± 0.09^{b} 28.38 ± 0.22^{c} 26.57 ± 1.09^{d} 14.61 ± 0.18^{a} 14.00 ± 0.15^{b}	Cooked Protein 14.27 ± 0.31^{a} 14.38 ± 0.64^{a} 11.56 ± 0.16^{c} 10.43 ± 0.49^{c} 21.01 ± 0.35^{a} 20.95 ± 0.16^{a}	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c} 5321.19 ± 441.57^{a} 2357.63 ± 61.70^{a} 2848.06 ± 50.60^{b}	$\begin{array}{c} \text{Springiness} \\ 0.93 \pm 0.02^{a} \\ 0.87 \pm 0.03^{b} \\ 0.86 \pm 0.03^{b} \\ 0.89 \pm 0.04^{c} \\ \end{array} \\ \begin{array}{c} 0.92 \pm 0.04^{a} \\ 0.86 \pm 0.01^{b} \\ \end{array}$	$\begin{array}{c} \text{Cohesiveness} \\ 0.56 \pm \ 0.01^{a} \\ 0.54 \pm \ 0.01^{b} \\ 0.56 \pm \ 0.01^{c} \\ 0.58 \pm \ 0.01^{d} \\ \end{array} \\ \begin{array}{c} 0.41 \pm \ 0.01^{a} \\ 0.53 \pm \ 0.02^{b} \end{array}$	$\begin{array}{c} \mbox{Chewiness} \\ 2792.53 \pm 385.95^a \\ 2325.82 \pm 229.15^b \\ 1238.74 \pm 167.99^c \\ 2776.96 \pm 264.46^a \\ 1312.78 \pm 63.33^a \\ 1300.45 \pm 69.16^b \\ \end{array}$	Resilience 0.41 ± 0.02^{a} 0.30 ± 0.01^{b} 0.35 ± 0.01^{c} 0.41 ± 0.01^{a} 0.20 ± 0.01^{a} 0.31 ± 0.02^{b}
Reduced Salt/Fat Sausage Sausage CoQ10 Commercial Sausage 1 Commercial Sausage 2 Reduced Salt/Fat Frankfurter Frankfurter CoQ10 Commercial Frankfurter 1	$\begin{array}{r} \mbox{Cooked Moisture} \\ 56.76 \pm 0.18^a \\ 50.85 \pm 0.11^b \\ 45.45 \pm 0.21^c \\ 33.01 \pm 1.43^d \\ \mbox{62.43 \pm 0.71}^a \\ 62.87 \pm 0.33^b \\ 56.32 \pm 0.01^c \\ \end{array}$	Cooked Fat 14.03 ± 0.17^{a} 18.38 ± 0.09^{b} 28.38 ± 0.22^{c} 26.57 ± 1.09^{d} 14.61 ± 0.18^{a} 14.00 ± 0.15^{b} 26.35 ± 0.19^{c}	$\begin{array}{c} \mbox{Cooked Protein} \\ 14.27 \pm 0.31^a \\ 14.38 \pm 0.64^a \\ 11.56 \pm 0.16^c \\ 10.43 \pm 0.49^c \\ \mbox{21.01} \pm 0.35^a \\ 20.95 \pm 0.16^a \\ 13.45 \pm 0.17^b \end{array}$	Hardness 5346.27 ± 499.91^{a} 5004.34 ± 312.41^{b} 2632.87 ± 508.11^{c} 5321.19 ± 441.57^{a} 2357.63 ± 61.70^{a} 2848.06 ± 50.60^{b} 3057.20 ± 69.68^{c}	$\begin{array}{c} \text{Springiness} \\ 0.93 \pm 0.02^{a} \\ 0.87 \pm 0.03^{b} \\ 0.86 \pm 0.03^{b} \\ 0.89 \pm 0.04^{c} \\ \end{array} \\ \begin{array}{c} 0.92 \pm 0.04^{a} \\ 0.86 \pm 0.01^{b} \\ 1.01 \pm 0.02^{c} \\ \end{array}$	$\begin{array}{c} \text{Cohesiveness} \\ 0.56 \pm \ 0.01^{a} \\ 0.54 \pm \ 0.01^{b} \\ 0.56 \pm \ 0.01^{c} \\ 0.58 \pm \ 0.01^{d} \\ \end{array} \\ \begin{array}{c} 0.41 \pm \ 0.01^{a} \\ 0.53 \pm \ 0.02^{b} \\ 0.58 \pm \ 0.01^{c} \\ \end{array}$	$\begin{tabular}{ c c c c c } \hline Chewiness \\ \hline $2792.53 \pm 385.95^a \\ \hline $2325.82 \pm 229.15^b \\ \hline $1238.74 \pm 167.99^c \\ \hline $2776.96 \pm 264.46^a \\ \hline $1312.78 \pm 63.33^a \\ \hline $1300.45 \pm 69.16^b \\ \hline $1798.59 \pm 24.98^c \\ \hline a \hline a \\ \hline a \hline a \\ \hline a \hline $	$\begin{tabular}{ c c c c c } \hline Resilience \\ \hline 0.41 \pm 0.02^a \\ \hline 0.30 \pm 0.01^b \\ \hline 0.35 \pm 0.01^c \\ \hline 0.41 \pm 0.01^a \\ \hline 0.20 \pm 0.01^a \\ \hline 0.31 \pm 0.02^b \\ \hline 0.43 \pm 0.01^c \\ \hline 0.43 \pm 0.01^c \\ \hline 0.01 \end{bmatrix} \end{tabular}$

Table 3. Physio-chemical Analysis Mean Values and Standard Deviation.

Values correspond to mean data, \pm corresponds to standard deviation. Note: a, b, c, d means with different letters in columns are significantly different (p < 0.05). Unshared *alphabetic superscripts* denote significantly different group means. E.g. Value ^a is the highest scoring attribute and would not be significantly different to another value with ^a.



Figure 1. ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for sausages.

Shown are the loadings of the X- and Y-variables for the first 2 PCs for \bullet = the individual treatments, \bullet = sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 26.63% and the calibrated variance was 26.92%.



Figure 2. ANOVA-Partial Least Squares Regression (APLSR) correlation loadings plot for Frankfurters.

Shown are the loadings of the X- and Y-variables for the first 2 PCs for \blacktriangle = the individual treatments, \bullet = sensory descriptor and instrumental variables. The validated explained variance for the model constructed was 18.84% and the calibrated variance was 19.16%.

Overall Discussion and Conclusion

The processed meat sector is a dynamic and ever changing part of the meat industry. Processed meats have always fulfilled a vital role by using the less commercial and usable parts of the carcass which in some cases would be seen as nothing more than waste and converting them into products that are enjoyed by consumers worldwide. They are often more convenient to eat than fresh meat as well as being cheaper and have developed in many countries as traditional dishes. The production of processed meat nowadays is driven by consumers demand for convenience, nutritional requirements, ethicality and safety.

This thesis began with the investigation of the European consumers' perception of processed meats and their viability as a functional food (Chapter 3). Over 60% of respondents perceived processed meats as an unhealthy product. The respondents in particular found the levels of salt and fat to be the greatest cause for health concerns from processed meat consumption. However, the majority of respondents in this study were female and previous studies have shown women to be more aware of potential health implications from meat consumption than men (Kubberod, Ueland, Rodbotten, Westad & Risvik, 2002). In fact the majority of men in this study were either unsure of the healthiness of processed meat or else rated them as healthy. Due to these gender differences the results from this study could have been different were equal male responses to female response obtained, in fact the percentage of respondents who dislike processed meats are likely to decrease significantly.

The respondents in the study showed positive feed-back in terms of bioactive compound addition in yogurt-style products, however the majority were found to be unsure of their feelings when used in meat-based products. This is likely due to a degree of unfamiliarity to meat products being marketed as functional and the pre-

conceived idea that processed meats are inherently unhealthy. When asked if they would consume meat-based functional food products the respondents answered in favour of the idea but were not willing to pay more for them.

Using the knowledge garnered from the preliminary survey study the next stage was decided as a look into salt and fat reduction in processed meats. Many of the recipes used by the meat industry were developed empirically over time rather than being grounded in a scientific methodology. Previous research has shown both changes in the consumers taste preference and also a more health conscious attitude towards meat purchase from the consumer (Resurreccion, 2003; Burton, Dorsett & Young, 1996; Young, 1996). Consequently it was postulated that a decrease in salt and fat levels could be obtained without the use of replacement ingredients. The products chosen varied in the degree that the meat was comminuted and consisted of; beef patties, pork breakfast sausages and beef and pork Frankfurters. The greatest hurdle to overcome in the development of reduced fat and salt products were the potential negative impacts to both the sensory and physical characteristics of the meat product as well as reduced shelf life or safety with respect to salt reduction. Therefore the main task of the subsequent three studies (Chapter 4, 5, 6) was to look at both the sensory and physiochemical effects from the reduction of salt and fat in these products and to try and pinpoint the most consumer acceptable formulations. The formulations used for each product type followed industry standard recipes modified slightly to adhere to a clean label approach. The effects of salt and fat reduction on the microbiological load of the meat products was not determined, as the focus of this thesis was directed towards sensory and functionality thresholds.

The study conducted on beef patties found that the fat and salt content had a major effect on a number of quality attributes. A full factorial design was found to be

the best option as it eliminated the potential of discrepancies in the data, especially for sensory evaluation. Twenty beef patties, varying in fat (30%, 40%, 50%, 60% w/w) and salt (0.5%, 0.75% 1.0% 1.25% 1.5% w/w) concentrations were produced, the average levels used in industry were found to be 50% fat and 1.5% salt. Each sample formulation was assessed by 25 regular consumers of meat product for sensory analysis, in duplicate. Every formulation also underwent instrumental testing for colour, moisture, fat, protein, ash, cooking loss and texture profile analysis.

The consumer sensory data showed that lowering the level of fat in beef patties produced products which were perceived as darker in appearance while low salt patties were found to be perceived as darker compared to higher salt patties. The decrease in both fat and salt also had significant effects on tenderness and mouth feel, producing products which rated higher in these attributes than patties containing more salt and fat. In terms of taste, the taste perception of salt was higher in high salt patties, and low salt patties were significantly negatively correlated to taste of salt. Off flavour was directly correlated to high fat patties.

Moisture levels were found to be correlated to lower fat and higher salt patties, while fat content was correlated to high fat and low salt patties. These impacted upon the level of cooking loss which was most significantly associated to higher fat patties and lower salt patties. Colour measurements showed significant correlations between L value and the highest fat patty both in a cooked and raw form, while a values were found to correlate to low fat patties due to the higher level of lean meat. Results from texture profile analysis showed correlations for hardness and cohesiveness to lower fat and higher salt patties. Higher fat and lower salt patties were also shown to be highly correlated to resilience and springiness.

From these data the most acceptable patty formulation was found to be 40% fat and 0.5% salt. This is a 20% decrease in fat and a 50% decrease in salt levels of average commercial patties and is within levels issued by governmental health authorities (FSAI, 2010).

Chapter 5 focused on the reduction of salt and fat levels in pork breakfast sausages. The formulations chosen were; 22.5%, 27.5%, 32.5%, 37.5% w/w for fat and 0.8%, 1%, 1.2%, 1.4%, 1.6%, 2%, 2.4% w/w for salt, these levels were chosen as the spanned incorporated the levels presently used in industry. Similar to the previous study on beef patties, each sample formulation was assessed by 25 regular consumers in duplicate for sensory analysis and also under went instrumental testing for colour, moisture, fat, protein, ash, cooking loss and texture profile analysis.

Lowering the fat content produced products which were rated to be lighter in appearance than higher fat samples, reductions in salt had a similar effect in producing lighter appearing products. Coarseness was found to be positively associated to low fat levels, while toughness were found to be correlated to higher fat products due to the increased lubrication provided by the extra fat. Salt reduction created products which the assessors described as more tender and coarse than higher salt formulations. Juiciness was reduced by the decrease in both salt and fat levels, these formulations scored significantly less in this attribute when compared with high fat and salt products.

For flavour and taste attributes, products containing more salt and fat were scored as having more of a salty taste. Meat flavour was directionally correlated to lower fat and salt formulations in comparison to those containing higher levels of fat and salt. There were no significant correlations observed for varying fat levels, however, high salt levels were found to have significant negative correlations to off-

flavour. The lower values for off-flavour in high salt samples may be due to the higher salt perception which could mask off-flavours present within samples.

From this study it was found that fat levels could be reduced in sausages without a significant reduction to product quality and overall acceptability. A reduction in salt levels were found to have a noticeable impact on consumer acceptability for sausages, By lowering the level of salt beyond 1.4% products were rated unfavourably, however, the salt levels can be lowered to a level that is recommended by the governmental heath agencies (FSAI, 2010). Therefore, the most acceptable and healthy breakfast sausage formulation was found to be 1.4% salt and 22.5% fat.

The reduction of salt and fat in pork and beef Frankfurters was explored in Chapter 6. The formulations chosen were; 10%, 15%, 20%, 25% w/w for fat and 1%, 1.5%, 2%, 2.5%, 3% w/w for salt, these levels were chosen as they reflect the levels presently used in industry. Just like the previous studies in Chapters 4 and 5, every individual Frankfurter formulation was assessed by 25 regular consumers in duplicate for sensory analysis and also under went instrumental testing for colour, moisture, fat, protein, ash, cooking loss and texture profile analysis.

Salt and fat were found to play a vital role in the both the sensory and physiochemical properties for Frankfurters. By lowering the salt and fat levels certain unfavourable changes occurred in certain aspects of Frankfurters quality. A reduction in fat and salt produced Frankfurters which panellists described as being darker in colour than higher fat and salt formulations. Toughness was not affected by fat concentration in the Frankfurters, however, formulations with lower salt levels were rated as having a more coarse mouthfeel and being texturally tougher. This reduction in textural quality was attributed to the increased level of cooking loss found in lower salt Frankfurters compared to their higher salt counterparts. Juiciness was also impacted by the reduction of salt and fat. The reduction in fat had negative effects on juiciness similar to findings for the beef patties (Chapter 4), higher fat formulations generally scored higher for juiciness due to the lubricating effects of fat in products (Mittal & Barbut, 1994 and Berry & Leddy, 1984). The reduction of salt causes less extraction of myofibrillar protein, these extracted proteins are the basis of the formation of the gel network which entraps water in meat products, thus making the products less juicy.

The taste and flavour of the Frankfurters were found to also be affected by a reduction in both fat and salt, with the perception of salt positively correlated to higher salt and higher fat formulations similar to pork breakfast sausages (Chapter 5). As with salt perception, meat flavour was found to be more associated with higher fat and salt formulations, this was likely due to the copious amounts of volatile compounds in animal fat (Brewer, 2012), and the flavour enhancing effects of salt (Silva, Morais, & Silvestre, 2003). However, those properties associated with fat and salt are also likely to contribute to the higher panellist scores for off-flavour in both higher fat and salt Frankfurters. Salt levels below 1.5% were shown to have a negative effect on consumer acceptability, with 2.5% salt concentrations being the most significantly preferred by consumers. No significantly preferred fat level was found, however, formulations containing the lower fat levels had directionally positive associations to acceptability. The optimum levels of salt and fat determined form this data was 2.5% salt and 10% fat, this level of salt is seen as acceptable in Frankfurter-style products by health authorities (FSAI, 2010).

The results from the Frankfurter trial showed the importance of the taste and textural properties of salt in the production of Frankfurters and a reduction in levels is

hard to achieve without using salt replacers. On the other hand fat levels could be potentially reduced without significantly affecting product quality and overall acceptability. In the end it was determined that formulations containing 15% and 10% fat with higher salt levels (2.5-3%) were significantly the most acceptable variants to consumers.

Supported with the information gleaned in Chapters 4, 5 and 6, the next stage of the thesis was the development of meat-based functional foods. As the focus of this thesis is on the direct fortification of processed meats with a bioactive, mid-processing was chosen as the method to develop a functional product. A micellerized form coenzyme Q10 (NovaSolQ[®]) was used as it was amphiphilic and provided the CoQ10 in a clear solution. The first task to assess the viability of the CoQ10 as a bioactive in meat was to test for bioaccessibility. Bioaccessibility is the quantity of a food constituent that is present in the gut, as a result of the release of this constituent from a food medium, and may be available to interact with an organism via intestinal absorption.

Chapter 7 focused on the effects of cooking and digestion on the concentration of CoQ10 in beef patties and pork sausages. The addition of micellarized CoQ10 was found to have a higher cooking retention level and digestibility in the pork sausages compared to beef patties, the difference however, was not statistically significant and was attributed to higher cooking loss in the patties. The post retention rate was found to be as high as 79% of the initially added CoQ10 which indicate that a sufficient level of CoQ10 survives the cooking process to have potential beneficial health effects in humans. Even more optimistic results were found in the digested samples where as much as 95 % of the CoQ10 that survived cooking would be available in the small intestine where absorption takes place. This study indicated the real potential that micellerized CoQ10 has as a functional ingredient in processed meat products.

With the positive results for CoQ10 as a functional ingredient in processed meat products (Chapter 7) the next and final stage of this thesis was to compare the CoQ10 enriched products and the reduced fat and salt products against pre-existing commercial products. Chapter 8 details the consumer sensory trial of reduced fat and salt beef patties and the beef patties enriched with CoQ10 against commercially available counterparts. This work also looked at the physiochemical properties of the CoQ10 enriched patties, which were found to behave identically to the beef patties not fortified with CoQ10.

Both products were found to have a better appearance compared to commercial products, but rated similarly in terms of tenderness, juiciness and flavour. The CoQ10 enriched patties, in particular, were found to be inconsequential in terms of impact on consumer sensory acceptance. The most significant change to samples was found to be an increase in yellowness (b* value), which was not perceived by consumers. In conclusion, the reduction of fat and salt in the patties produced products which consumers found to be just as acceptable as commercially available products. Similarly the reduced salt/fat patties fortified with CoQ10 were also found to be just as acceptable commercially available products.

Chapter 9 shows the findings from the consumer sensory trial of reduced fat and salt pork breakfast sausages and Frankfurters with and without CoQ10 fortification against commercially available counterparts. Similarly to Chapter 8 the physiochemical properties of the CoQ10 enriched products were also investigated. Both the CoQ10 enriched sausages and Frankfurters were found to behave identically to the products not fortified with CoQ10 in terms of texture, cooking loss and

composition, the only difference found was in the yellowness (b* value) like the patties in Chapter 8.

The reduction of fat and salt in sausages was shown to produce products which consumers found to be as acceptable as commercially available products. The CoQ10 fortified sausages, like the fortified beef patties, had no significant negative impact on consumer acceptance levels. The reduced fat Frankfurters were found to be less acceptable to consumers than commercially available higher fat equivalents, however, no differences were found when compared to the fortified Frankfurters. This indicates that there is a potential for the addition of CoQ10 to an existing commercial brand of Frankfurter without negatively impacting product quality. Overall, the studies from Chapter 8 and 9 indicate the strong potential for micellerized CoQ10 in a range of processed meat products.

Conclusion

In summary, the research findings from this thesis indicate that the potential to reduce salt and fat in beef patties and pork breakfast sausages can produce products which are acceptable to consumers. Differences were found between the three differently comminuted meat systems. Both salt and fat reduction were shown not to negatively impact on consumer acceptability in patties and sausages, while only fat reduction could be achieved in Frankfurters without negative consumer feedback. These findings provide both industry and regulatory agencies with the knowledge that fat and salt reduction is possible in processed meats, but limits exist to the extent of this reduction. This can help push the meat industry to produce processed meat products which have improved nutrional profiles in terms of health while maintaining consumer acceptance. The results from Chapter 3, provide positive information regarding the use of bioactives in processed meat products to produce functional foodbased meat products. The exploration of CoQ10 addition is detailed in Chapters 7, 8 and 9 and found that processed meat products can be enriched with micellerized CoQ10 which is both bio-accessible and does not negatively affect consumer acceptability of sensory qualities of the product. Based on the findings of this thesis the ability to create meat based functional foods without negatively affecting consumer acceptability can be achieved and is a viable means to improve the nutritional quality of processed meats and therefore the health of those who regularly consume these products.

Future Work

The results from this Thesis will be used to develop a follow-on project looking at the development of other reduced fat and salt processed meat products such as cured and uncured meats. This project will investigate the reduction and or replacement of salt and fat with respect to functionality, food safety, consumer sensory quality and commercial viability. The minimum concentrations of preservatives will also be identified while maintaining the above attributes in order to determine the very limits of such removal. This project has been titled as Development of Consumer Accepted Low Salt and Low Fat Irish Traditional Processed Meats: PROSSLOW. It will focus on traditional Irish processed meat products. Sensory consumer research will be employed to optimise each of these approaches as well as using active coatings on packaging innovation, through the use of non contact bioactive materials, to synergistically replace preservatives have been removed, reduced or replaced. The project will show clear quantitative goals for the sequential reduction of salt and fat in traditional Irish processed meat products.
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