# Innovative Food Product Development using Molecular Gastronomy: a Focus on Flavour and Sensory Evaluation 

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# Innovative Food Product Development using <br> Molecular Gastronomy; A Focus on Flavour and <br> <br> Sensory Evaluation 

 <br> <br> Sensory Evaluation}

THESIS SUBMITTED TO DUBLIN INSTITUTE OF TECHNOLOGY IN FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY Mark Traynor, BA School of Culinary Arts and Food Technology,<br>College of Arts and Tourism,<br>Dublin Institute of Technology.

Supervisors: Dr. Róisín Burke \& Dr. Catherine Barry-Ryan


#### Abstract

The primary goal of this research was to develop novel ice cream products using the principles of molecular gastronomy. An ice cream model system (emulsion) was developed, in which the effects of ingredient levels on stability and formation were investigated and optimised using Response Surface Methodology (RSM). Two characteristic volatiles of banana (isoamyl acetate and furfuryl acetate) were added to the optimised emulsion, and their headspace emission was quantified using Solid Phase Microextraction with Gas Chromatography Mass Spectrometry. RSM was used to investigate the main and interactive effects of ingredient, salt and pH levels on the headspace emission of these volatiles. Salt was found to significantly influence ( $\mathrm{p} \leq 0.05$ ) headspace emission of isoamyl acetate.

The pairing of novel foods (banana and bacon (B+BN), banana and basmati rice $(\mathrm{B}+\mathrm{R})$, and banana and olive oil ( $\mathrm{B}+\mathrm{O})$ ) was investigated as an important sensory phenomenon with a key interest in determining how different components in the selected food pairings (both volatile and non-volatile) affect and interact with other components to influence sensory perception. Consumer sensory evaluation $(\mathrm{n}=85)$ showed that $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$ were significantly more acceptable ( $\mathrm{p} \leq 0.05$ ) pairings than $\mathrm{B}+\mathrm{O}$. Correlation of descriptive sensory analysis $(\mathrm{n}=28)$ and organic volatile profiling was conducted to try to elucidate the hedonic results. Two ice cream product recipes were formulated to provide a matrix for the two preferred novel food pairings ( $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$ ). Product acceptability was assessed using two consumer panels representing general and specific markets. Significant differences ( $\mathrm{p} \leq 0.05$ ) between hedonic results of the two panels suggest that the $\mathrm{B}+\mathrm{R}$ ice cream may be more suited to a general retail product, whereas the $\mathrm{B}+\mathrm{BN}$ ice cream may be more suited to a selective culinary market.


## DECLARATION

I certify that this thesis which I now submit for examination for the award of Doctor of Philosophy, is entirely my own work and has not been taken from the work of others, save and to the extent that such work has been cited and acknowledged within the text of my work.

This thesis was prepared according to the regulations for postgraduate study by research of the Dublin Institute of Technology and has not been submitted in whole or in part for another award in any Institute or University. The work reported in this thesis conforms to the principles and requirements of the Institute's guidelines for ethics in research.

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Signature $\qquad$ Date $\qquad$

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## List of abbreviations

| A | Absorbance |
| :---: | :---: |
| AAS | Absorption spectrometry |
| ANOVA | Analysis of variance |
| AOAC | Association of Official Analytical Chemists |
| APLSR | ANOVA-partial least squares regression |
| Asym | Final emulsion droplet diameter |
| $\mathrm{a}_{\text {w }}$ | Water activity |
| B | Banana |
| BN | Bacon |
| B+BN | Banana and bacon food pairing |
| $B+\mathrm{R}$ | Banana and basmati rice food pairing |
| $\mathrm{B}+\mathrm{O}$ | Banana and extra virgin olive oil food pairing |
| CA | California |
| CAR | Carboxen |
| CCF | Central Composite Faced Centred experimental design |
| cm | Centimetre |
| C5 | Carbon number 5 |
| $\mathrm{C}_{6}$ | Carbon number 6 |
| C9 | Carbon number 9 |
| DIT | Dublin Institute of Technology |
| DDW | Distilled deionized water |


| DS | Mean emulsion droplet diameter |
| :---: | :---: |
| DVB | Divinylbenzene |
| $\mathrm{D}^{32}$ | Sauter number |
| e | Slope |
| EU | European Union |
| eV | Electron volt |
| FAO | Food and Agricultural Organisation |
| FCP | Free Choice Profiling |
| FEMA | Flavour and Extract Manufacturers Association |
| FDA | Food and Drugs Administration |
| $F$-value | Fisher test value |
| g | Gram |
| $\mathrm{g} / 100 \mathrm{~g}$ | Grams per 100 grams |
| g/ Kg | Grams per kilogram |
| GC/MS | Gas chromatography mass spectrometry |
| GMF | Genetically modified food |
| GPA | Generalised Procrustes Analysis |
| GRAS | Generally recognised as safe |
| H | Height |
| HC | Height of cream layer in emulsion |
| HE | Initial emulsion height in the tube |
| HS | Height of sedimentation phase in emulsion |
| HS-SPME | Headspace solid-phase microextraction |


| $\mathrm{HNO}_{3}$ | Nitric acid |
| :---: | :---: |
| Inc. | Incorporated |
| INRA | French National Institute of Agricultural Research |
| ISO | International Organisation for Standardisation |
| k | Number of factors in RSM |
| 1 | Path length |
| L | Litre |
| LLE | Liquid-liquid extraction |
| lrc | Natural logarithm transformation of first order rate constant |
| LSD | Fishers Least Squared Difference |
| Ltd. | Limited |
| M | Molar |
| m | Metre |
| MA | Massachusetts |
| MCO | Multi criterion optimisation |
| mg | Milligram |
| ml | Millilitre |
| mL /minute | Millilitres per minute |
| mm | Millimetre |
| MSG | Monosodium glutamate |
| n | Number of subjects |
| NaCl | Sodium chloride |


| NaOH | Sodium hydroxide |
| :---: | :---: |
| NC | North Carolina |
| NIST | National Institute of Standards and Technology |
| Nls | Nonlinear least squares |
| O | Extra virgin olive oil |
| OVAT | One variable at a time |
| O.D. | Container diameter |
| $P$ | Probability |
| PA | Pennsylvania |
| PDMS | Polydimethylsiloxane |
| pH | Power of hydrogen |
| pKa | Acid dissociation constant |
| plc | Private limited company |
| ppb | Parts per billion |
| ppm | Parts per million |
| PTFE | Polytetrafluoroethylene |
| QDA | Quantitative Descriptive Analysis |
| R | Basmati rice |
| $R^{2}$ | Co-efficient of determination |
| $R_{0}$ | Initial emulsion droplet diameter |
| RMSE | Root mean square error |
| rpm | Revolutions per minute |
| RSD | Residual standard deviation |


| RSM | Response surface methodology |
| :---: | :---: |
| s | Seconds |
| SE | Standard error |
| SD | Standard deviation |
| SPME | Solid-phase microextraction |
| SR | Emulsion stability ratio |
| SS | Sum square error |
| $\mathrm{s}^{-1}$ | Unit of shear rate measurement |
| TIC | Total ion chromatogram |
| UK | United Kingdom |
| USA | Unites States of America |
| v/v | Volume per volume percentage |
| W | Watts |
| w/v | Weight (mass) per volume |
| $\alpha$ | Alpha |
| $\beta$ | Beta |
| $\chi^{2}$ | Chi square |
| \% | Percentage |
| - $\alpha$ | Low setting for variables in experimental design |
| $+\alpha$ | High setting for variables in experimental design |
| $\alpha$ | Medium setting for variables in experimental design |
| ${ }^{\circ} \mathrm{C}$ | Degrees Celsius |
| $\mu \mathrm{g}$ | Microgram |

$\varepsilon$

2-AP
$<$
$\leq$
$>$

Error term

2-acetyl-1-pyrroline

Less than

Less than or equal to

Greater than

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## Chapter 1

### 1.1 General introduction

Few things are as intrinsically intertwined with humanity as food, which supports life and underpins social relations. Science explores the world and the mechanism of natural phenomena, specific scientific methods of investigation are used to examine every aspect of the environment. Cooking being such an imperative part of our world, draws on scientific principles (chemical actions and physical reactions), and therefore deserves specific scientific studies (Blumenthal, 2008; This, 2005a). Molecular gastronomy is the scientific discipline dedicated to the exploration and investigation of culinary mechanisms of phenomena which are related to the sensory perception of food (Snitkjær, 2010). While, molecular gastronomy is still relatively novel, it is an important development in terms of modern gastronomy. Although molecular gastronomy arrived in Ireland considerably later than in other countries (France, Spain, Portugal, etc.), it is now rapidly developing and has great potential for the progress of Ireland's culinary industry and it's gastronomic tourism (Valverde et al., 2011). The principles of molecular gastronomy (physics and chemistry) can provide a blueprint for "high tech" product development (Rodgers, 2011).

The food industry is one of the most important industries in the European Union, which spans a wide range of economic activities, with a high relevance for employment and economic output (Wijnands et al., 2007). According to CSO (2012), the food industry accounts for a substantial part of the Irish economy, contributing an annual output of approximately $€ 20$ billion. The development of a food and agriculture sector which is sustainable from an economic and social point of view is critically important to Ireland's future development (DAFM, 2013). Food product development is necessary for survival of the product in a competitive global market (Wang \& Lin, 2009). New and alternative food processing methods, as well as novel combinations of existing
methods (sous vide cooking etc.), are continually being sought by industry in the pursuit of producing better quality foods (Rastogi, 2010).

Chefs, culinary experts and food processors have become interested in the intelligent design of foods with optimal flavour characteristics (Roberts \& Taylor, 2000). The modern kitchen is a meeting place where chefs, who are normally characterised by their artistry, creativity, and craft, can interact with scientists who are normally characterised by their empiricism, rationality and adherence to the scientific method (van der Linden et al., 2008). The application of the principles of molecular gastronomy to the production of food products has great potential for providing customers with novel and exciting food products, while supplying food manufacturers with competitive advantages and unique selling points.

### 1.2 Molecular gastronomy

Molecular gastronomy utilises scientific methods of investigation to better understand and control the molecular, physicochemical and structural changes that occur in foods during their preparation and consumption, with a focus on sociology and enjoyment of food (Barham et al., 2010; van der Linden et al., 2008). It can be considered a subfield of food science, with an aim of understanding culinary transformation in the restaurant or domestic kitchen (Snitkjaer, 2010). Chefs struggle to discover methods that guarantee a perfect outcome for a specific recipe, molecular gastronomists strive to find perfection for all recipes using scientific methods of investigation (Blanck, 2007). Hence, the identification of optimum methods of creating foods with pleasurable flavour and texture is a key component of molecular gastronomy (This, 2005a).

### 1.2.1 Background and history of molecular gastronomy

In 1988, Nicholas Kurti, the low-temperature physics Professor at Oxford University (UK), and the French chemist Hervé This co-founded the academic discipline of molecular gastronomy (This \& Rutledge, 2009). Molecular gastronomy was introduced based on the observation that food science had slowly drifted, over many decades, towards the investigation and preservation of large scale industrial processes and production, nutrition, food safety and food composition (Barham et al., 2010). Conversely, such rational insights had not been deployed in the small scale culinary realms of the restaurant and home (de Solier, 2010).

There has been a lot of misunderstanding and miscommunication by the public, the media, professional chefs and scientists regarding the true meaning of molecular gastronomy (Snitkjaer, 2010). The term "molecular gastronomy" has been misused to describe a style of cooking which is often more correctly referred to as "molecular cooking" (van der Linden et al., 2008). Molecular cooking differs from earlier styles of cooking (haute and nouvelle cuisine) through a reliance on food science and technology principles, practices and techniques in the kitchen (Vega \& Ubbink, 2008). Molecular cooking is the application of science to technique, and involves applying the results and knowledge obtained from molecular gastronomy, and for this reason, molecular gastronomy is not a style of cooking (van der Linden et al., 2008).

### 1.2.2 Molecular gastronomy research and application

For the continuous progression of molecular gastronomy as a scientific discipline, research within the scientific scope of molecular gastronomy is imperative. A large scale three year (2002 to 2005) collaborative project called the Introduction of Innovative Technologies in Modern Gastronomy for Modernisation of Cooking (INICON) was largely funded by the European Union (Blanck, 2011). INICON has promoted collaboration between European chefs, scientists, companies and culinary schools through this European technology transfer programme (Snitkjaer, 2010). This project combined the efforts of chefs at four restaurants (Au Crocodile, el Bulli, The Fat Duck, and Grashoff), researchers at educational and research and development institutions (Ecoles Gregoire-Ferrandi, the Molecular Gastronomy Working Group at the French National Institute of Agricultural Research (INRA), and TTZ-BremerhavenBIONARD), and members of the food industry (Alpha-Tec, Cosmos Armatica Internacional, and Iberagar) (Blanck, 2011).

In addition to this large scale collaborative project, numerous smaller scale yet equally noteworthy studies have concentrated on a wide range of important physical and chemical culinary phenomena. In particular, recent studies on meat stock have focused on the preparation (This et al., 2006), the flavour development (Snitkjaer, et al., 2010) and the effects of wine on meat stock (Snitkjaer et al., 2011). Another area that has been a key focus of scientific investigation in recent times is low temperature vacuum cooking (sous vide cooking). In particular, the influence of sous vide cooking on meat texture (Roldán et al., 2013; Christensen et al., 2012; del Pulgar et al., 2012; Mortensen et al., 2012; Pakula \& Stamminger, 2012). The modifications induced by various culinary and industrial treatments on pigment systems of immature pods of green beans (Phaseolus vulgaris L.) has also been explored (Valverde \& This, 2008; Valverde et al.,
2007). While, the principals of the phenomena food/flavour pairing have likewise been an emphasis of scientific investigation in the field of molecular gastronomy (Traynor et al., 2013; Ahn et al., 2011; Kort et al., 2010).

### 1.3 Dispersion science

### 1.3.1 Food dispersions

Food materials are on the whole highly non-homogeneous systems. Very few foods can be characterised as being a single phase, in fact the vast majority of foods are of a multiphasic nature (Barham et al., 2010). Foods are often characterised by the simultaneous presence of a diverse range of components dispersed in mixed solutions (Ettelaie, 2003). Hence, food products are generally complex chemical and physical systems, typically existing as dispersed systems, formally known as colloids (Dickinson, 2006). For this reason, dispersion science is of great important and interest to molecular gastronomists. Disperse systems can be described in terms of a number of dispersed phases (mesoscale particulate structures), surrounded by a continuous phase or matrix (Barham et al., 2010).

The dispersed phases can derive from natural food products such as globular proteins in milk, or be artificially created via food processing such as oil droplets in mayonnaise (Aguilera \& Stanley, 1999). Next to these mesoscale structures of the dispersed phase, food contains smaller molecular species, like salts, sugars, polyols and phospholipids, which moderate the properties of the continuous or dispersed phases, or their interfaces by serving as surface active molecules (surfactants), plasticisers and humectants (van der Sman \& van der Goot, 2009). In general, depending on the properties of the continuous and dispersed phases, relatively simple food dispersions
can be classified into the categories which are outlined in Table 1.1. In order to optimise the textural and shelf life characteristics of food, it is clear that is important to have an understanding of colloid science concepts and techniques (Dickinson, 2002). The molecular gastronomist and food scientist strives to understand how specific components (proteins, lipids and carbohydrates) can control the stability and rheology of complex colloidal systems.

Table 1.1 Dispersed system classification with food examples.

| Common name | Disperse phase | Continuous phase | Examples |
| :---: | :---: | :---: | :---: |
| Foam | Gas | Liquid | Whipped cream |
| Solid foam | Gas | Solid | Bread, meringue |
| Emulsion | Liquid | Liquid | Milk, mayonnaise |
| Gel | Liquid | Solid | Jelly |
| Sol | Solid | Liquid | Egg white |
| Aerosol | Solid/liquid | Gas | Flavoured "Airs" |
| Solid dispersion | Solid | Solid | Chocolate |
| Powder | Solid | Gas | Flour |

Source: Adapted from Gaman \& Sherrington (1996).

### 1.3.2 Food emulsions

An important class of food dispersions are emulsions (Schramm, 2005). Traditionally an emulsion can be defined as a dispersion of droplets of one liquid in another, the two being immiscible (McClements, 2005). The culinary industry is one of many industries that heavily rely on the use of emulsions and emulsifiers (Guzey \& McClements, 2006). Emulsions play an important role in the formulation of foods; many natural and processed foods exist either partly or wholly as emulsions (salad dressings, mayonnaise, cream liqueurs, etc.) or have been in an emulsified state at some time during their existence (breads, soft drinks, etc.) (Charcosset, 2009; Dalgleish,
2006). Emulsions can also be ingredients which participate in the formation of more complex products such as yoghurts, ice creams and whipped products (Leal-Calderon et al., 2007).

From a physicochemical point of view, emulsions are thermodynamically unstable systems, rapidly or slowly separating into two immiscible phases over a period of time (Borwankar et al., 1992). Emulsions have several distinctive properties which stem partly from the fact that the dispersed phase is usually less dense than the continuous phase, so that droplets rise (or cream) in a gravitational field (Robins et al., 2002). Illustrated in Figure 1.1 are the most common processes of emulsion destabilisation; droplet-droplet coalescence, flocculation, creaming and sedimentation (Tcholakova et al., 2006).


Figure 1.1 Schematic diagram of common instability mechanisms that occur in food emulsions: creaming, sedimentation, flocculation, coalescence, Ostwald ripening and phase inversion.

Source: Piorkowski \& McClements (2013).

These destabilisation processes greatly influence shelf life and texture of emulsions (Dickinson \& McClements, 1995; Phillips et al., 1994). A major coalescence mechanism which leads to a gradual coarsening of emulsion droplets is Ostwald ripening, which by and large has a detrimental effect on the shelf life of these types of food products (Ettelaie, 2003). The flocculation of emulsions is commonly via the depletion interactions, which are important in many food systems (Manoj et al., 2000). Depletion flocculation is an entropic effective attraction in which non-adsorbed species (polysaccharide molecules, protein aggregates and surfactant micelles) are excluded from a small lens-shaped region between two droplets (Dickinson, 2002). The osmotic pressure due to the surfactant molecules in the continuous phase exerts a pressure driving the droplets together (Figure 1.2) (Robins et al., 2002).


Figure 1.2 Schematic diagram of depletion flocculation.
Source: adapted from Koltay \& Feke (1999).

Emulsion stability refers to the ability of an emulsion to resist changes in its properties over time: the more stable the emulsion, the more slowly its properties
change (McClements, 2005). The perceived quality of emulsion based food products is strongly influenced by their stability, rheology and appearance (Mirhosseini et al., 2008a). A main indicator of loss of stability is an increase in emulsion droplet mean diameter, and the growth rate of the droplets can reveal the mechanism responsible (Silva et al., 2010). Enhancing an emulsion based product's resistance to destabilisation can be done by reducing the droplet size (McClements, 2005).

Das \& Kinsella (1990) explain that forming a kinetically stable emulsion can be achieved through the addition of surfactants. Surfactants such as monoglycerides and phospholipids are common molecules in foods because they are used to stabilise interfaces. They can be charged or neutral, but in both cases the enthalpy of mixing of their hydrophilic and the hydrophobic parts is very high, which confers on them their typical amphiphilic nature (Mezzenga et al., 2005). Emulsifiers are surfactants which lower surface tension and prevent droplet flocculation by absorption on the droplet surfaces (Krstonosic et al., 2009). Lecithin is a small molecule surfactant which is one of the most commonly used emulsifiers in the culinary industry (Whitehurst, 2004). Legislators classify it as generally recognised as safe (GRAS) and it is accepted as a natural ingredient by consumers (Bylaite et al., 2001).

Polysaccharides are surfactants employed as thickeners of emulsions which are commonly added to the aqueous phase of oil in water emulsions to confer long term emulsion stability (Quintana et al., 2002). Viscosity modification or gelation in the aqueous continuous phase is the main stabilisation action (Dickinson, 2003). Xanthan gum has the ability to increase the viscosity of the aqueous continuous phase at relatively low concentrations, therefore it is one of the most employed thickeners to stabilise dispersed oil droplets in food emulsions such as pourable salad dressings and sauces (Hemar et al., 2001).

### 1.3.3 Ice cream as a complex dispersed system

Ice cream is an example of a complex food system (Scholten \& Peters, 2012). The particles making up ice cream can have a variety of forms (Figure 1.3), they might consist of emulsion droplets, small gas bubbles (air), ice crystals, self-assembled solidlike aggregates of proteins and fats, as encountered with casein micelles, or even comprise of just single globular protein macromolecules dispersed in an unfrozen sugar solution (Ettelaie, 2003). Thus, ice cream would be more accurately described as a "multiple suspension/foam/emulsion" (This, 2007).


Figure 1.3 The structure of ice cream viewed by thin section transmission electron microscopy.
a: air bubble.
f: fat globule.
fc: fat cluster.
Source: Goff (2002).

The manufacturing of ice cream usually begins by formulating, pasteurising, homogenising and cooling an emulsion premix, aging of the emulsion is then followed
by aerating and freezing under high shear conditions in a scraped surface freezer (Schmidt, 2004; Segall \& Goff, 2002). In terms of formulation, ice cream is essentially milk, cream, water, and sugar (Scholten \& Peters, 2012). The key to making a good ice cream is to formulate a mix that will freeze into a balanced structure of ice crystals, concentrated cream, and air (McGee, 2004). Several of the processing steps contribute to fat structure development in ice cream (Goff, 1997). Homogenisation of the emulsion premix converts the bulk fat into finely divided droplets (Patmore et al., 2003).

Two important processes take place during aging. First, the emulsifiers adsorb to the surface of the fat droplets, replacing some of the milk protein (Figure 1.4). This produces a weak membrane that is strong enough to stabilise the emulsion under the static conditions in the aging tank, but makes the emulsion unstable under shear force. Second, the fat inside the droplets begins to crystallise. It is essential that aging is long enough for crystallisation to occur and for emulsifiers to displace some of the protein since both of these processes are important precursors to the next stage in ice cream production (Clarke, 2004).

The aeration and freezing process involves numerous physical changes. Firstly, gas bubbles are introduced into the sweetened cream mixture through churning, whipping and kneading (Dondurma, 2012). The gas phase volume varies greatly from a high of greater than $50 \%$ to a low of $10-15 \%$. Air is distributed in the form of numerous small air bubbles of size range 20-50 $\mu \mathrm{m}$ (Goff, 2002). Proteins and surfactants form and stabilise the foam phase, while partial coalescence of the fat emulsion causes both absorption of fat at the air interface and formation of fat globule clusters that stabilise the lamellae between air bubbles (Patmore et al., 2003).


Figure 1.4 Fat droplet during ice cream aging process, showing adsorption of milk proteins and emulsifiers at the surface of the fat droplet and crystallisation of the fat. Source: Clarke (2004).

During the freezing stage the individual fat globules are partially coalesced into a three-dimensional network (Berger, 1997). The presence of some fat crystals is critical for fat structure development as protruding crystals may initiate the interaction between droplets, thus, helping to hold flocculated fat together (van Boekel \& Walstra, 1981). Sugars have two major functions in ice cream; they make it taste sweet, and they control the amount of ice formed and hence the softness of ice cream (the higher the ice content, the harder the ice cream) (Clarke, 2004). When sugar is added to water, the temperature at which water freezes will be less than $0^{\circ} \mathrm{C}$, which is known as freezing point depression. Upon freezing, water is removed from the solution as ice, effectively increasing the sugar concentration in the unfrozen phase (Scholten \& Peters, 2012).

### 1.4 Food system and process investigation through response surface methodology

The complex colloidal and polymeric nature of food systems makes their investigation more difficult (Mezzenga et al., 2005). To allow for reliable interpretation of the experimental data, the use of model systems remains essential in order to gain quantitative knowledge (Dickinson, 2002). Traditionally, optimisation in analytical chemistry has been carried out by monitoring the influence of one variable at a time (OVAT) on an experimental response (Bezerra et al., 2008). The major disadvantage of this method is that it does not include the interactive effects among the variables studied, subsequently the complete effects of the parameter on the response are not depicted (Lundstedt et al., 1998).

Response Surface Methods (RSM) are designs and models based on the fit of a polynomial equation to a set of experimental data for working with continuous treatments when finding the optima or describing the response is a goal (Oehlert, 2000). Another goal of RSM is to understand how the response changes in a given direction by adjusting the design variables (Bradley, 2007). Toward these objectives, linear (main) or squared polynomial (quadratic) functions are employed to describe the system studied and, consequently, to explore (modelling and displacing) experimental conditions until its optimisation (Teófilo \& Ferreira, 2006). Linear relationship between independent and dependent variables can be defined by a linear function (first-order models), while, quadratic relationships with curvature in the response surfaces require higher degree polynomial (second or third-order models) to be employed. In general, all RSM problems use either one or the mixture of the both of these models (Bradley, 2007).

In addition to analysing the effects of the independent variables, RSM generates a mathematical model which describes the chemical or biochemical processes (Anjum et al., 1997). The visualisation of the predicted model equation can be obtained by the response surface three-dimensional plot and contour two dimensional plot (Baş \& Boyacı, 2007). First-order models are appropriate for describing a flat surface with or without tilted surfaces, and are not suitable for analysing maximum or minimum points (Montgomery, 2005). The main advantage of RSM over OVAT is the reduced number of experimental trials needed to evaluate multiple parameters and to optimise a process (Mirhosseini et al., 2008b). RSM also takes into account the interactions among variables unlike OVAT (Leardi, 2009). Thus, RSM is an effective statistical technique for optimising complex processes because it allows more efficient and easier arrangement and interpretation of experimental conditions compared to OVAT (Gan \& Latiff, 2011).

Many culinary processes and mechanisms have been studied using RSM for optimisation and investigation of variable interactions, especially emulsion and flavour release. Buffo et al. (2002) studied the influence of time and temperature on the emulsifying properties of gum acacia in oil in water emulsions using RSM. Gharibzahedi et al. (2012) modelled the optimisation of formulation variables and physical stability of oil in water emulsions using RSM. While, the optimisation of headspace concentration of emulsions (Mirhosseini et al., 2009a; Mirhosseini \& Tan, 2009) and soursop (Cheong et al., 2011) were also modelled using RSM. Advances in statistics and computing enable a variety of modelling applications such as temperate distribution in ovens to predict the rate of Maillard reactions (Rodgers, 2008). The prediction of the rate of such reactions offers a powerful quantitative tool in terms of food product development (Martins et al., 2001).

### 1.5 Sensory perception of food

The primary purpose of perception is to seek out objects in our environment, particularly those that are biologically important (Prescott, 2012). Humans are decisively influenced by their sense of taste and odour, therefore, human history is closely tied to the development and usage of flavours (Zieglar \& Zieglar, 1998). Sensory science itself is a relatively young discipline and has only been in existence for roughly 60 years (Drake, 2007). For a large portion of that time, research on sensory perception of food has primarily concentrated on taste, flavour, and some simple mechanical properties of a product such as viscosity, firmness, and hardness.

The International Organisation for Standardisation (ISO) defines flavour as $a$ complex combination of the olfactory, gustatory and trigeminal sensations perceived during tasting, the flavour may be influenced by tactile, thermal, painful and/or kinaesthetic effects (ISO 5492, 2009). Flavour is considered one of the most important attributes determining the acceptance of food by the consumer (Tromelin et al., 2006). Flavour is an interaction of the food and the consumer, not a property of the food alone, so no study of flavour is complete unless the consumer is considered as well as the chemistry and physics of the food (Piggot, 2000). Although flavour is initially influenced by the receptors in the eyes, nose, tongue and mouth lining, it is the brain which interprets the overall sensation occurring in the mouth (Taylor \& Hort, 2004).
1.5.1 The major senses involved in the sensory evaluation of foods

During gustation, food is first brought into the mouth and subsequently processed in the oral cavity (Chen, 2009). Taste buds on the tongue, palate, soft palate, and areas in the upper throat (pharynx and laryngopharynx) located within three types of papillae (fungiform, foliate, and circumvallate papillae) detect the taste sensations of bitter, salty, sour, sweet and umami (Figure 1.5) (Barham et al., 2010). In addition to these sensations, there are a variety of chemical sensations known as the trigeminal sense (chemesthesis) (Meilgaard et al., 2007). When chemical irritants such as ammonia, ginger, horseradish, onion, chilli pepper, menthol stimulate these trigeminal nerves in the mucosa of the eyes, nose, and mouth to give hot, burning, tingling, cooling or astringent sensations (Kemp et al., 2009). Trigeminal neurons are buried below the surface in the mouth, for this reason the response to stimuli is slow in onset and long lasting (Reineccius, 2006). For example, contrary to the sense of taste, which is most intense for the few seconds the food is in the mouth, pungency typically lasts for minutes to tens of minutes (Barham et al., 2010).

The tongue and the oral cavity are also sensitive to size, shape, texture, consistency, and temperature of a food object (Auvray \& Spence, 2008). Textural stimuli originate from the structure and mechanical properties of food and the way these break down during masticating (Taylor \& Hort, 2004). The detection of sensations related to contact/touch (force, particle size and heat) by tactile receptors of the lips, tongue and surfaces of the oral cavity is known as somesthesis (Kemp et al., 2009). Kinesthesis perceptions correspond to the mechanical movement of muscles as a result from stress exerted by the muscle of the jaw or tongue (hardness, stickiness, etc.), and the sensations of the resulting strain (compression, shear, etc.) within the food being masticated (Meilgaard et al., 2007).

## buds located



Figure 1.5 The taste receptor fields of the human oral cavity. (a) The tongue, showing the four different types of papillae. (b) A fungiform papilla containing taste buds. (c) Cross section of a taste bud showing the taste pore. (d) The taste cell. (e) Receptor sites for bitter, sweet, sour, salty, and umami substances.

Source: adapted from Goldstein (2009).

The largest contribution to the diversity of food flavours comes from their volatile or airborne molecules (Lawless \& Heymann, 2010). It is estimated that up to $80 \%$ of the information about our food depends on olfaction (Blake, 2004). To be perceptible, aroma compounds must be volatile and released into the air and transferred
to the olfactory cells located in the olfactory epithelium at the roof of the nasal cavity, which is directly linked to the olfactory bulb of the brain (de Roos, 2006; Pernollet et al., 2006). Stimulation of the olfactory receptors can be achieved by two distinct routes (Figure 1.6): either via the nose by orthonasal olfaction (sniffing), or by retronasal olfaction via the mouth, during eating and drinking, as volatile chemicals rise up through the nasopharynx (Auvray \& Spence, 2008).


Figure 1.6 Olfactory perception via orthonasal route and retronasal routes.
Source: adapted from Goldstein (2009).

The perceived appearance of food can have a profound impact on expectations and actual taste and flavour sensory characteristics (Wei et al., 2012). The total appearance of an object is formed by interaction of the visual appearance properties (colour, translucency, gloss, and surface texture properties) with the human response (Caivano \& del Pilar Buera, 2012). Such sensory characteristics may include sweetness,
pureness, refreshing, freshness, naturalness, flavour intensity, thirst-quenching, and liking (Zellner \& Durlach, 2002).

### 1.5.2 Multisensory flavour perception

Human perception of food flavour and texture during consumption is a complicated process in which taste, mouth feel, vision, olfaction, the trigeminal system and auditory signals contribute to the total appreciation of a food product (Visschers et al., 2006). Sensory attributes cannot be related to a single physical property of the food (van Vliet et al., 2009), and are perceived when each of the human sensory receptor organs interact with the physicochemical properties of the food (Kemp et al., 2009). Traditionally, the senses involved with the perception of flavour in food were considered to be limited to olfaction, taste, and the somatosenses (irritation, tactile, and thermal). However, it is now acknowledged that numerous other sensory inputs are processed by the brain to result in flavour perception (Reineccius, 2006). As shown in Figure 1.7, the multiplicity of interactions between taste, smell, touch, sound, vision and the trigeminal system has led numerous researchers to propose flavour as the term for the combinations of these systems, unified by the act of eating (Small \& Prescott, 2005; Abdi, 2002; Prescott, 1999; McBurney, 1986).

There is no combination of sensory modalities that excludes taste and smell and still creates a flavour. Colour, texture, sound, irritation and temperature have all been definitively demonstrated to influence flavour, either through a perceptual interaction or a physical one (Delwiche, 2004). The simple notion that flavour involves a wide range of the well-known taste and aroma modalities has been replaced by a realisation that it is a multi-modal phenomenon (Taylor \& Hort, 2004). Consequently, flavour perception
should be used as a term to describe the combinations of taste, smell, the trigeminal system, and touch, to which visual and auditory cues should be added (Auvray \& Spence, 2008).


Figure 1.7 Summary of perceptual interactions evoked during ingestion.
Source: adapted from Delwiche (2004).
Arrowhead indicates a modality that has been demonstrated to interact with another modality.

The study of the multisensory processes involved in flavour perception is important for a better understanding of the processes used by consumers to assess the acceptability and flavour of new products (Shepherd, 2006; Blake, 2004; Gilbert \& Firestein, 2002). A complete flavour experience depends on the combined responses of the senses and the cognitive processing of these inputs (Auvray \& Spence, 2008); the
release of flavour chemicals in the mouth, the transport processes to the receptors, the specificity and characteristics of the receptors, the transduction mechanisms and the subsequent processing of signals locally and at higher centres in the brain (Taylor \& Hort, 2004).

### 1.5.3 Food sensory analysis

Sensory or organoleptic evaluation of food has been conducted for as long as man has evaluated the goodness and badness of food and water (Meilgaard, et al., 2007). Sensory evaluation can be seen as a link between research and development, specifically technical aspects of food, and consumer and marketing research, focusing on consumers' behaviour and psychology (Dijksterhuis, 1997). The sensory attributes of food are not necessarily the parameters leading to food acceptance or rejection, but rather the assessor's preference for particular levels of the food attributes (Shepherd, 1990).

In the food industry, sensory analysis is the food company's single most important analytical tool and an asset for research and development. In particular, combining both sensory analysis and chemical analysis (flavour analysis) can be a powerful tool for resolving many types of difficult flavour problems (Kemp et al., 2009). Sensory and instrumental analysis of food has been successfully employed in tandem for flavour profiling in tomatoes (Auerswald et al., 1999), fruit smoothies (Keenan et al., 2012) and blackberries (Du et al., 2010).

### 1.5.3.1 Consumer tests

Consumer tests (affective tests) have been proven to be highly effective as a tool used to design food products that will sell in larger quantities or command higher prices (Meilgaard et al., 2007). Consumer testing assesses subjective responses to a product using both qualitative and quantitative methodologies. Researchers can gain an insight into consumer preferences, attitudes, opinions, behaviours and perceptions concerning food products (Kemp et al., 2009). A consumer acceptance test is a panel test usually involving only 50 to a 100 panellists, and is conducted throughout the different phases in the product development process. There are two approaches to consumer sensory acceptance testing: the measurement of preference and the measurement of acceptance. Acceptance tests measure consumer acceptance or liking (hedonic rating) of a product (Moskowitz et al., 2012). The consumer acts as a measuring instrument, assigning numbers or words from a scale. In contrast, preference tests measure the appeal of one food or food product over another (Stone \& Sidel, 2004).

### 1.5.3.2 Descriptive analysis

Information obtained from the description of the sensory characteristics of food and beverages enable companies to make more informed business decisions (Stone \& Sidel, 2004). Sensory profiling is increasingly viewed as a way to explain and possibly anticipate consumer preferences, and is usually carried out by conventional profiling techniques, such as Quantitative Descriptive Analysis (QDA) (Delarue \& Sieffermann, 2004). QDA can be time consuming and expensive due to the need to develop an agreed vocabulary and to carefully and extensively train the assessors. Free Choice Profiling
(FCP) was developed to circumvent some of these problems with the conventional profiling techniques (Lachnit et al., 2003).

The main benefit of FCP methods is their rapidity and ease of use (Delarue \& Sieffermann, 2004). Another advantage of FCP methods over conventional descriptive methods is that a defined descriptive vocabulary isn't required, therefore, untrained consumers may be used as descriptive assessors (Delahunty et al., 1997). This relatively recent methodology is attractive because it does not demand a training stage and individual sessions are possible (Albert et al., 2011). FCP methods have been previously utilised to describe different foods such as red fruit jams (Dairou \& Siefffermann, 2002), dairy products (Delarue \& Siefffermann, 2004), commercial apple and pear purées (Tarea, et al., 2007), flavour perception of bread odour (Lassoued, et al., 2008; Poinot et al., 2007), jellies (Blancher et al., 2007) and wines (Perrin et al., 2008).

### 1.5.4 Hedonic reactions to unfamiliar foods

A novel product refers to a newly developed product unfamiliar to the consumers (Stolzenbach et al., 2011). The reluctance to consume or the tendency to avoid or dislike an unfamiliar food is a hallmark of omnivores known as food neophobia (Henriques et al., 2009; Martins \& Pliner, 2006). On the other hand, food neophilia is the urge towards novelty, with a tendency to try new and unfamiliar foods (van Trijp \& van Kleef, 2008). However, people can be also in the middle of continuum, being neutral between neophilia and neophobia (Asperin et al., 2011).

There are many factors that influence food neophobia, including; sociodemographic characteristics such as culture, age, gender, education, economic and social environments (Olabi, et al., 2009; Flight, et al., 2003). The common interest among culinary researchers and professionals would be to understand and reduce the level of food neophobia in a targeted population (Asperin et al., 2011). Generally, exposure, familiarity, nutritional information, and positive experiences increase food neophilia and decrease neophobia (McFarlane \& Pliner, 1997). For example, the more exposed an individual is to diverse cultures, the more likely he/she will be more neophilic (Tourila, et al., 2001).

A framework for understanding acceptance and rejection of both familiar and unfamiliar foods comes from a taxonomy of basic motivational dimensions identified by Fallon and Rozin (1983). Each dimension is bipolar, with food near the negative poles being rejected and foods near the positive poles being accepted (Martins \& Pliner, 2006). The three basic motivational dimensions underlying reactions to a food substance are identified by Martins et al. (1997) as: (1) sensory-affective (liking or disliking the taste, smell or appearance of a food), (2) anticipated consequences (expectations about the beneficial or harmful consequences of ingestion), and (3) ideational (knowledge of the nature or origin of the substance which can lead to it being positively transvalued or considered disgusting). As a consequence, an unfamiliar food that does not fall into one's acceptable category will be rejected (Dovey et al., 2008).

### 1.6 Flavour chemistry

### 1.6.1 Organic volatile compound emission from food

Release of organic volatile compounds from foods is an essential requirement for flavour perception (Roberts \& Taylor, 2000). Knowledge of volatile-food and volatile-volatile interactions is important for understanding flavour perception (Plug \& Haring, 1993). Flavour compounds are volatile and odorous at atmospheric pressure, their retention is a function of the availability of the flavour compounds in the gas phase and, therefore, of the affinity of these compounds for the food matrix (Voilley \& Souchon, 2006). The perception of the aroma compound at the level of the olfactory epithelium partly depends on its availability in the vapour phase (Seurve, 2007). This is dependent on the nature and concentration of volatile compounds present in the food, as well on as their availability for perception as a result of interactions between the food matrix components and the aroma compounds in the food (Bakker, 1995). The solubility of flavour compounds in the different phases (aqueous or lipid) of the foods has a significant role in the behaviour of the flavours in the matrix and will have an overriding influence on their sensory perception (Landy et al., 1998).

Flavoursome foods can often contain hundreds of organic volatile compounds, with interactions between these compounds being complex (Chung et al., 2003). For example, a mixture of two volatile compounds usually elicits a weaker aroma than the sum of its parts, the perceived intensity of flavour compounds having a logarithmic rather than a linear relationship with concentration (Wright, 2010). Other complex volatile-volatile and volatile-non-volatile (taste) interactions exist. Some taste compounds (such as Monosodium glutamate (MSG)) can increase the perceived aroma
intensity of foods, while the perceived intensity of tastes may also be altered by the aroma of volatile compounds (Salles, 2006).

Any type of interaction between a flavour compound and a food constituent which results in a restriction of the movement of a flavour stimulus to a sensory receptor will ultimately influence flavour perception (Reineccius, 2006). Also, these volatiles are not uniformly released from the food, but commonly interact with macronutrients such as proteins, fats and carbohydrates, which can result in an uneven release from the food (Chung et al., 2006). For this reason, food matrix components can bind, entrap or encapsulate volatile flavour compounds resulting in a reduction in the rate of flavour release and flavour intensity (Naknean \& Meenune, 2010). Such binding phenomena generally involve interactions that are specific to the flavour and composition of the food (Malone \& Appelqvist, 2003). The interactions between flavour compounds and the three major food components (lipids, proteins and carbohydrates) are summarised in Table 1.2.

Physical interactions are determined by the air-food partition coefficient of the aroma compound, which is a measure for the volatility of the aroma compound in the food medium, and also by the mass transfer coefficient determining the kinetics of the aroma release (de Roos, 2006). Numerous factors influence the flavour release from food matrices, including chemical interactions (hydrogen, ionic or covalent bonding) between food and the flavouring, physical considerations (physical barriers to release) as well as human factors such as number of teeth, chewing efficiency, chewing time and breathing process (Reineccius, 2006). The interactions between flavour substances and major food components can be attractive or repulsive interactions. Attractive interactions involve fixation of flavour compounds on food components, whereas
repulsive interactions concern the release of aroma compounds (van Ruth \& Roozen, 2010).

Table 1.2 Types of interactions that may occur between flavour compounds and major food components.

| Food component | Possible Interactions |
| :---: | :--- |
| Lipids | solution |
|  | dispersion |
|  | adsorption |
|  | entrapment |
| Proteins | adsorption |
|  | absorption |
|  | specific binding |
|  | covalent interactions |
|  | entrapment |
|  | encapsulation |
| Carbohydrates | adsorption |
|  | entrapment |
|  | complexation |
|  | encapsulation |
|  | viscosity/diffusion limitations |

Source: Kinsella (1989).

### 1.6.1.1 Volatile-lipid interactions

It is generally acknowledged that flavour-lipid interactions are one of the most important factors to affect the release and perception of aroma compounds, having the biggest effect on the partitioning of flavour compounds between products and the gaseous phase (Ollivon, 2006). The majority of flavour compounds are hydrophobic and therefore favourably partition into the lipid phase rather than into the aqueous or gas
phases, which can alter the balance of the perceived aroma (Wright, 2010). The overriding effect is the role of lipid as a flavour solvent. Lipids can adsorb and solubilise hydrophobic flavour compounds (Reineccius, 2006). Lipophilic flavour compounds bind to the lipid molecules by weak, reversible van der Waals forces (Plug \& Haring, 1993). The generally hydrophobic nature of flavour compounds results in considerable differences in headspace composition, higher amounts of lipids generally reduce the volatility of hydrophobic odorants (Kerŝiene et al., 2008). Whereas, in the absence of lipids, retention of lipophilic flavours is poor, resulting in high headspace concentrations (Plug \& Haring, 1993). In numerous model emulsions and more complete foods, the differences in headspace concentration and the resulting sensory intensity due to modifications of the lipid level are well noted (Doyen et al., 2001; Welty et al., 2001; Gwartney et al., 2000; Haahr et al., 2000; Brauss, et al., 1999).

### 1.6.1.2 Volatile-carbohydrate interactions

The chemical properties of carbohydrates will determine chemical reactions with flavour compounds, while the physical properties will determine their influence on flavour compound mass transport both in the food and from the food (Reineccius, 2006). Some carbohydrates can bind volatiles via hydrogen bonding between appropriate functional groups, others, such as starch, consist of three-dimensional structures with hydrophobic regions capable of forming inclusion complexes with various hydrophobic volatiles (Godshall \& Solms, 1992). Hydrocolloids not only modify viscosity and consistency, but often reduce intensities of odour, taste and flavour (Delwiche, 2004). This retention of the aroma compound in a food matrix can be influenced by a decrease of diffusion of the aroma compounds due to an increase of viscosity and by the presence of interactions between polysaccharides and volatile
compounds (Seurve et al., 2007). At lower water content, carbohydrates of various molecular weights may form a glassy matrix which has excellent retention properties for aroma compounds (Delarue \& Giampaoli, 2006).

### 1.6.1.3 Volatile-protein interactions

Two types of interactions can occur between flavour compounds and proteins: (1) reversible physical adsorption via non-covalent interaction and (2) irreversible chemical reaction via covalent linkages (van Ruth \& Roozen, 2010). These interactions are influenced by the amount of protein and amino acid composition, types of flavouring components, presence of other food components, ionic strength (salts), pH (ionic form and conformation), temperature and time (Fischer \& Widder, 1997). Bonds between aroma compounds and proteins are generally weak and include reversible bonds, van der Waals bonds, hydrogen bonds, and/or hydrophobic interactions (Kerŝiene et al., 2008). Chemical interactions and mass transfer effects are diverse due to the wide range of chemical structures available for interaction such as amino acid chains and hydrophobic pockets, the viscosity and gel structures in proteins (Reineccius, 2006).

### 1.6.1.4 Physicochemical interactions

The physicochemical behaviour of flavour compounds in food matrices is one of the most important parameters involved in their activity and sensory perception. Hence, this behaviour can have pronounced effects on the flavour quality of foods, particularly emulsions (Mirhosseini et al., 2008a). Physicochemical interactions take place between
aroma compounds and food constituents, which modify the partition of these organic volatile compounds inside the food, thereby influencing the retention and/or the release of the aroma compounds (Guichard, 2002).

The perception of flavour and aroma volatiles in foods is influenced by the solubility of volatile compounds, which subsequently can be affected by the level of salt $(\mathrm{NaCl})$ present in a food (Mitchell et al., 2011). Salt concentrations will have a significant influence on the ionic strength of a food, which will in turn influence the solubility of flavour compounds depending on their hydrophobicity (Rabe et al., 2003). The practice referred to as "salting out", is a well-known technique for increasing headspace concentrations of volatile molecules prior to separation and quantification by headspace gas chromatography (Flores et al., 2007; Pérez-Juan et al., 2007). These compounds bind considerable amounts of water to build hydration shells during solubilisation, resulting in reduced water activity ( $\mathrm{a}_{\mathrm{w}}$ ) due to the formation of strong ion-dipole interactions between the salt ions and water (Rabe et al., 2003). This fashions an increased flavour release from food due to the decreased availability of water molecules for the solubilisation of flavour compounds (Flores et al., 2007).

When added to aqueous solutions, mono- and di-saccharides interact with water molecules, producing a "salting out effect" (Delarue \& Giampaoli, 2006), whereby the sugar interacts with water, increasing the concentration of flavour compounds in the remaining volume of free water (Friel et al., 2000). It has been found that pH has an impact on the release of some aroma chemicals. At a pH below the acidity constant
 thereby less soluble in the aqueous phase. This would drive the acid into the headspace, increasing its contribution to the aroma profile (Reineccius, 2006). In contrast, odorants
in the ionized form below their pKa , e.g. amines or pyrazines, are more soluble in water and therefore exhibit less volatility (Bortnowska, 2010).

### 1.6.2 Flavour volatile profiles of foods

Approximately 12,000 volatile compounds have been identified in foods (Grosch, 2001), with some individual foods and beverages such as coffee, chocolate and bread containing up to 1,000 volatile compounds (Cheetham, 2010). Of these volatile compounds identified in foods, it is approximated that only around $5 \%$ play a significant role in the formation of the characteristic aroma of a specific food (Mistry et al., 1997). These unique chemical substances are known as "character impact compounds" and contribute the recognisable sensory impression for the particular flavour or aroma even at low concentrations levels. Examples of which are benzaldehyde in cherry, vanillin in vanilla and diacetyl in butter (McGorrin, 2012).

In order for an aroma to be active (i.e. be detected by the olfactory cells in the epithelium), the concentration of the molecule has to be above the specific aroma threshold for that volatile compound, which varies greatly from molecule to molecule (Jeleń et al., 2012). The intensity of a flavour is roughly related to its concentration in the food divided by its threshold for perception (Cheetham, 2010). Flavour quality and character are known to change with concentration (Wright, 2010). Fisher \& Scott (1997) explained that a chemical such as trans-non-2-enal has more than one recognition threshold. It possesses a woody character just above its detection threshold of 0.1 ppb . However, above 8 ppb it elicits fatty notes, and becomes unpleasant at 30 ppb , while at 1000 ppb it has a strong flavour of cucumber.

### 1.6.2.1 Flavour volatile profile of banana

Many studies have focused on the volatiles in fresh banana (Mayr et al., 2003; Nogueira et al., 2003; Liu \& Yang, 2002; Jordán et al., 2001; Ibanez et al., 1998). More than 250 organic volatile compounds have been identified in banana (Jayanty et al., 2002). Characteristic volatile esters, alcohols, acids and carbonyls are the four main compound classes that give strength and character to banana aroma, while amines and phenols also contribute (Mayr et al., 2003). In particular, esters represent the more numerous compounds in banana (Jordán et al., 2001), and have been attributed as being the main contributors to the characteristic fruity banana notes (Imahori et al., 2013; Vermeir et al., 2009).

Acetates have been noted as being of particular importance due to their high concentrations and low odour thresholds (Pontes et al., 2012). Specifically, amyl esters have been accredited with eliciting banana notes, while butyl esters contribute the fruity notes (Liu \& Yang, 2002). Isoamyl acetate and isobutyl acetate have been identified as being the two most important character impact compounds of banana aroma (Mayr, 2003). Both molecules elicit banana, pear and fruity sensory notes, while isobutyl acetate also has a slight rum note. Other compounds which elicite typical banana aroma notes are hexanal, 3-hydroxy-2-butanone, 2-pentanone, 2-pentanol, 3-methyl-1-butanol and eugenol (Vermier, 2009). Isoamyl alcohol, isobutyl butyrate, butyl butyrate, ethyl isovalerate, and ethanol have been identified as major volatile compounds of banana aroma (Liu \& Yang, 2002).

### 1.6.2.2 Flavour volatile profile of basmati rice

Studies of rice revealed that among 200 identified organic volatile compounds, only a few contribute to it's characteristic aroma (Jezussek et al., 2002; Widjaja et al., 1996). The characteristic impact compound responsible for the aroma of rice was reported as 2-acetyl-1-pyrroline (2-AP), a low odour threshold lipophilic compound contributing to a popcorn-like aroma (Yoshihashi, 2002). Yang et al. (2008) found that aldehydes and aromatic compounds were the most abundant odour-active compounds in basmati rice. They listed hexanal, 1-pentanol, ( $E$ )-2-hexenal, p-xylene, 2-heptanone, heptanal, 2-acetyl-1-pyrroline, ( $E$ )-2-heptenal, benzaldehyde, 1-octen-3-ol, 2pentylfuran, octanal, 3-octen-2-one, ( $E$ )-2-octenal, guaiacol, 2-nonanone, nonanal, p-menthane-3-one, $(E)$-2-nonenal, naphthalene, dodecane, decanal, $(E, E)$-2,4 nonadienal, (E)-2-decenal, and ( $E, E$ )-2,4-decadienal as being the most potent odourants in basmati rice. In addition, alcohols were found to represent $14.89 \%$ of the total aroma of basmati rice (Paule \& Powers, 1989). Hexanol was the major compound detected for this class of constituents, followed by oct-1-en-3-ol, reported as having a characteristic mushroom-like aroma (Buttery \& Kamm, 1980). Benzyl alcohol, 3-methyl butanol and 2-phenylethanol have also been detected in relatively high amounts in basmati rice (Tava \& Bocchi, 1999).

### 1.6.2.3 Flavour volatile profile of extra virgin olive oil

The aroma of extra virgin olive oil is composed of over 100 organic volatile compounds, most of which are present in very low concentrations (Contini \& Esti, 2006). $C_{5}, C_{6}$ and $C_{9}$ saturated and unsaturated aldehydes and alcohols and their esters are substances that mainly characterise virgin olive oil aroma (Cavalli et al., 2004;

Morales et al., 1995). The "cut grass aromas" of virgin olive oils have been attributed to several compounds such as nonanal, hexanal (Baccouri et al., 2007), (Z)-3-hexenal, (Z)-3-hexenyl acetate, hexyl acetate and (Z)-3-hexen-1-ol. So called "green aromas" have been attributed to ( $E$ )-2-hexenal, $(E)$-3-hexen-1-ol, $(E)$-2-hexen-1-ol (Angerosa et al., 2000; Aparicio et al., 2000), and 3 pentanone (Morales \& Tsimidou, 2000). Servili et al. (1997) found that (E,E)-2-4-heptadienal and ethylbenzaldhyde correlated with the "green apple" sensory perception in virgin olive oils, while 1-octanol and methyl salicylate were associated with "fatty" sensory perceptions. The hot spicy flavour of virgin olive oils has been previously attributed to the active compound pinene (Ekundayo et al., 1988). Acetic acid, ethanol and ethyl acetate have all been associated with "vinegar" or "wine" flavours in olive oils (Kalua et al., 2007).

### 1.6.2.4 Flavour volatile profile of bacon

Approximately 150 organic volatile compounds have been identified in the headspace volatiles of fried bacon. Of these compounds, hydrocarbons, aldehydes, ketones and alcohols have been reported in the headspace, while small amounts of pyrazines, furans, pyridines and other nitrogen and sulphur compounds were also present (Ramarathnam \& Rubin, 1994). Pyrazines such as 2-ethyl-5-methylpyrazine and 2-ethyl-3, 5-dimethylpyrazine have been found to contribute to the characteristic bacon flavour. Some furans and pyridines such as 2-pentylfuran and 3, 4-dimethylpyridine have been described as meaty aroma contributors to bacon flavour (Timón et al., 2004). Sulphur compounds have been noted as being important contributors to meat flavour in cooked foods due to their low flavour threshold (Flores et al., 1997). Yu et al. (2008) detected thioethers and thiol compounds in traditional Chinese bacon, which included dimethyl disulfide, 3-(methylthio) propanal, dimethyl trisulfide and methanethiol. These
compounds are considered to be particularly important aroma compounds of bacon produced through the Maillard reaction during cooking (Mottram \& Nobrega, 2002). Furans and thiophenes with a thiol group in the 3-position, and related disulfides, have been shown to possess strong meat-like aromas and exceptionally low odour threshold values (Mottram, 1998). Other thiols and disulfides, containing, 2-furanylmethyl moieties have also been found in the volatiles of heated meat systems (Madruga \& Mottram, 1995). The aldehyde produced via Strecker degradation 3-methylbutanal has been found to be a main contributor to the flavour of bacon (Ho et al., 1983).

### 1.6.3 Volatile analysis of foods

Many methods have been used for flavour volatile analysis. The most typically utilised methods for extraction and preconcentration are headspace techniques, purge-and-trap, liquid-liquid extraction, and simultaneous distillation and extraction (Liu \& Yang, 2002). Most of the traditional analytical sample preparation techniques, such as liquid-liquid extraction (LLE) rely on the exhaustive removal of analyte from the sample matrix by the use of a large volume of extraction phase (Vuckovic et al., 2010). This can be time consuming, labour-intensive, involve errors and losses in each stage, while waste disposal of solvents adds extra costs and creates health hazards (Vas \& Vékey, 2004).

### 1.6.3.1 Headspace solid-phase microextraction (HS-SPME)

In foods, headspace analysis is one of the options for instrumental determination of volatile compounds in a sample as the headspace contains all the volatiles that are responsible for the odour sensation (Chambers \& Koppel, 2013). Headspace sampling
techniques are frequently used in the determination of the volatile compounds present in foods (Zhao et al., 2010). A very successful approach to sample preparation for headspace analysis is solid-phase microextraction (SPME), invented by Pawliszyn and co-workers in 1989 (Arthur \& Pawliszyn, 1990). SPME is a rapid sampling technique where volatiles interact with a fibre-coated probe inserted into the headspace (Berna et al., 2005). In comparison to the traditional analytical sample preparation techniques, SPME is an equilibrium sample preparation technique, where only a small amount of extraction phase is used so that only a small portion of the analyte is removed from the sample (Vuckovic et al., 2010). SPME is a sensitive, selective and inexpensive solvent free method, providing better precision and linearity response with improved detection limits, which can also save preparation time while being easy to handle (Kataoka et al., 2000; Marsili, 1999; Zhang \& Pawliszyn, 1993). SPME has been routinely used in combination with gas chromatography mass spectrometry (GC/MS) and successfully applied to a wide variety of compounds, especially for the extraction of volatile and semi-volatile organic compounds from food samples (Vas \& Vékey, 2004). GC/MS itself is an excellent method for quantification and identification of aroma compounds (Berna et al., 2005) and is the single most widely used technique in flavour studies (Reineccius, 2010).

Figure 1.8 depicts the key component of a SPME device, which is a fused-silica fibre that is coated on the outside with an appropriate stationary phase such as polydimethylsiloxane (PDMS) (Yu et al., 2008). There are two typical SPME applications, sampling gases (headspace SPME (HS-SPME)) or sampling solutions. In both cases the SPME needle is inserted into the appropriate position through a septum, the needle protecting the fibre is retracted and the fibre is exposed to the environment (Ulrich, 2000). Analytes establish equilibria among the sample matrix, the headspace
above the sample, and a stationary phase coated on a fused silica fibre (Mirhosseini et al., 2008b). The technique is based on a sorption procedure (absorption and/or adsorption, depending on the fibre coating) (Arthur et al., 1992). After sampling, the fibre (Figure 1.9) is retracted into the metal needle (for mechanical protection), and the next step is to thermally desorb the analyte from the fibre to a capillary column of the chromatograph (Vas \& Vékey, 2004).


Figure 1.8 Schematic diagram of a commercial solid-phase microextraction device.

Source: Vas \& Vékey (2004).

The choice of the fibre coating is a key factor, since the type and amount of compounds that are extracted from the sample depends on the physical and chemical characteristics of the fibre stationary phase and on the film thickness (Yu et al., 2008). The affinity of the fibre for an analyte relies on the principle of 'like dissolves like', and coating fibres having different properties or thicknesses are selected in accordance with target compounds (Kataoka et al., 2000). HS-SPME has been successfully applied to the
analysis of flavour in foodstuffs such as oils (Cavalli et al., 2004), fish (Duflos et al., 2005), cooked turkey (Brunton et al., 2000), beef (Moon \& Li-Chan, 2004), cheeses (Costa et al., 2010), and banana (Pontes et al., 2012).


Figure 1.9 Headspace solid-phase microextraction gas chromatography mass spectrometry procedure.

Source: adapted from Vas \& Vékey (2004).

### 1.7 Food pairing

One approach to new product development is the pairing of novel foods or flavours. Food pairing (or flavour pairing) is the creative or artsy side of cooking and searching for and grasping successful flavour pairings has naturally been on the mind of many a chef (de Klepper, 2011). Food pairing is the coupling of flavours to produce an experience that is more appreciated than either of the two flavours alone (Møller, 2013). Food pairing has for the most part been described for the combinations of wines with foods, with most wine label giving menu suggestions (Kort et al., 2010). However, in more recent times, hypotheses regarding flavour pairings have been put forward. Two
food pairing online software tools for research and development support are currently available; Foodpairing (foodpairing.com, SenseforTaste, Bruges, Belgium) and Flavorstudio (senspirellc.com, Senspire, CA, USA).

The concept that foods can combine well when they share major flavour components is one such hypothesis that has been put forward in recent years (Møller, 2013). This hypothesis was first appreciated by the Firmenich scientist François Benzi in 1992, resulting in the creation of the online software tool Foodpairing.com. This hypothesis has received considerable attention over the past decade among chefs and food scientists (Ahn et al., 2011). This food pairing tool is based on the flavour pairing hypothesis, and is aimed at providing research and development support for professional chefs and bartenders. Users of the software tool can graphically view the similarities between food ingredients and food products on a visually interactive "food pairing tree" (Figure 1.10) (SenseForTaste, 2013).


Figure 1.10 Food pairing tree.
Source: Foodpairing.com (SenseforTaste, Bruges, Belgium).

Flavorstudio is a food pairing software tool for research and development support. This software utilises an alternative hypothesis of flavour pairing; a mathematical algorithm is used to identify patterns from a large database of over 1 million recipes to suggest ingredients that may pair well together. This particular software focuses on historical and geographical gastronomic patterns of flavour pairings as opposed to a purely chemical approach. Users can manually adjust the mathematical algorithm on a slider scale to reflect the strength of the relationship between ingredients (Figure 1.11). However, it must always be remembered that actual perception of flavour quality, character and intensity is very subjective and very dependent on prior consumer experience, so that large regional, cultural and age differences exist, even for some quite basic flavour sensations (Wright, 2010).


Figure 1.11 Flavorstudio online software food pairing tool working screen.
Source: senspirellc.com (Senspire, CA, USA).

### 1.8 Food Innovation

### 1.8.1 Innovative food product development

Customers are more demanding and more engaged in the food product development process and their interests in more differentiated and exciting food products have been observed in recent years (Stolzenbach et al., 2013; Topp, 2007). It is clear that a change in the mode of operation of product developers from supplyorientated activities to demand-orientated activities has developed (van Boekel \& Linnemann, 2011). Developing new and innovative products grants producers the capability to compete in mature and developed markets. Furthermore, these products perform several roles: give some protection against price competition, replace products that face declining sales at the end of their lifecycles, and contribute to creating customer satisfaction and loyalty (Grunert et al., 2004).

The key to market success lies in a balance between responsiveness and proactiveness and in short-term success from incremental innovation and long-term success from more radical new innovation (van Trijp \& van Kleef, 2008). In an increasingly globalising food market, innovation is an essential strategic tool for food enterprises to achieve competitive advantage, to stand out from competitors and to fulfil consumer expectations (Wang \& Lin, 2009; Gellynck et al., 2007; Menrad, 2004). However, the more a food product is differentiated, the more choices have to be made by product developers, and these choices must be in line with consumer preferences (Grunert et al., 2011). Thus, successful development of innovative food products requires an excellent understanding of consumers' perceptions, expectations and attitudes towards food products (Linnemann et al., 2006). For this reason, consumer integration in food innovation and development activities can increase the amount of diversified and
customer based knowledge obtained, which helps to reduce the innovation failure rate (Song et al., 2013). Moreover, customer integration also helps to create innovative ideas and feedback regarding concepts or prototypes for new products (Prahalad \& Ramaswamy, 2004).

It is commonly acknowledged that risks and difficulties are intrinsic in new product development (Hoyer et al., 2010). Food innovations are often rejected by consumers as a result of food neophobia (Barrena \& Sánchez, 2013). Nevertheless, the arrival of new products in the marketplace has created a climate of ambivalence or insecurity, in which some innovations meet opposition and suspicion (Grunert \& Valli, 2001). Many technology-based innovations (e.g., information technology) have been incorporated into daily life with high levels of consumer acceptance, on the other hand, others have been met with substantial resistance such as genetically modified foods (GMFs) and food irradiation (Ronteltap et al., 2007). As a result, $80 \%$ to $90 \%$ of all newly developed products in the food and beverage market fail (Duber-Smith \& Black, 2012).

### 1.8.2 The importance of food innovation to the Irish economy

The considerable development in research and food innovation has led to an increase of new foods entering the market and which have a key role to play in the sustainable development and competitiveness of the sector (Bäckström et al., 2004). Ireland is striving to create a world-class research system that drives innovation and economic success (DAFM, 2013). The agriculture and food sectors are extremely important to the Irish economy. One in seven jobs in Ireland is in the food industry, with over 300,000 people working in the growing processing and selling of Irish food
(Phelan \& O'Connell, 2011). It is a well-known fact that high levels of investment in research and innovation is a major driver of economic growth (Grunert et al., 2008). In particular, science, technology and innovation are vital to the Irish economic and social progress. The development of the 'smart' or innovation-based economy is the key challenge facing Ireland, even within the largely uncharted territory of the current financial crisis (DETE, 2009). For Ireland to become one of the most highly innovative food countries in the world, Irish food exporters should focus on innovation leading to brand building based around customer feedback as a means to capturing greater market value (Bord Bia, 2010). Some steps have already been taken to begin this process. Kerry Group have begun the construction of a global flagship technology and innovation research and development centre in Naas, Co. Kildare, which will be operational by 2015 (The Daily Business Post, 2012).

### 1.8.3 Gastronomy as a driver for food innovation

Gastronomy and the industry of fine dining are becoming major drivers for food innovation (Aguilera, 2009). Food manufacturers assess marketplace trends, and very often monitor what trends are currently popular in restaurants in order to develop concepts for new products (Moskowitz et al., 2009). Chefs are integral in the process of creating novel food products through actively participating in the generation of ideas, process development, and final applications (Valdovinos, 2009). For many years now, chefs have experimented with liquid nitrogen, rotary steamers, thermo-regulators, ultrasonic mixers and other laboratory type equipment to design radically different food items such as vodka mayonnaise, transparent pasta stuffed with caviar and others (Villariano, 2006). Product development centres of food multinationals are listening to
chefs and bringing them into their quarters to profit from their approaches, thereby increasing the dissemination, popularity and use of new products, and subsequently expanding niches for their products (Valdovinos, 2009). Food product developers should be alert about this top-down trend of innovation as it may lead to improved goods with added novelty (Aguilera, 2009). Furthermore, the use of analytical methods combined with the principles of molecular gastronomy and supported by statistical predictive models such as response surface methodology can produce outstanding outcomes in food product development (Rodgers, 2008).

### 1.9 Proposed study

### 1.9.1 Motivation

A proactive approach to the modern consumer's fundamental desires and needs, where food is appreciated for it's source of enjoyment and pleasure and not solely for nutrition and sustenance is required for survival and progression in the current economic climate. Traditional methods of new product development (especially in large companies) involve the coordination and cooperation of numerous departments (food engineering, processing, marketing etc.) at different steps and stages throughout the development process. Furthermore, technical knowledge and information obtained through separate scientific experimental investigation of culinary phenomena can also be applied (usually carried out by a separate party). As molecular gastronomy is a science, with an aim of obtaining new knowledge through scientific investigation, the incorporation of a molecular gastronomy approach to new product development would allow for the direct technical application of newly obtained knowledge. Thus, this intelligent design of food begins at a molecular level, investigating the chemical and
physical interactions between food constituents themselves, and also between the consumer and the food itself.

Furthermore, food product developers are typically skilled or have experience in specific areas of the food product development process. These areas are typically either scientific based (food engineering, food processing, nutrition and diet, food safety etc.) or culinary based areas (butchery, charcuterie, garde manger, bakery etc.). With a broader skill set (both culinary and scientific), molecular gastronomists are scientists who apply their knowledge to culinary products. This knowledge can then be applied to improve the quality of the gatronomique experience of food products for the retail sector. Hence, a major advantage of having a molecular gastronomist involved in the product development process is more efficient communication through having a greater understanding of both culinary and scientific terminology and specific processes. For this reason, molecular gastronomists can communicate with all the separate departments involved in the development process, helping to break down the communication barriers between science and culinary arts.

Therefore, a molecular gastronomy approach allows for a more holistic understanding of food products, providing insight on how to manipulate and optimise new food products to maximise the gastronomic experience of the consumer. Additionally, a consumer integrated product development approach offers an insight into the consumer's perception of the products. Developing novel food products using qualitative and quantitative methods of scientific investigation in this thesis will ultimately provide the consumer with high quality novel foods.

### 1.9.2 Research process

Ice cream products were selected as the emulsion based food products to be developed in this thesis as they are widely consumed (Clarke, 2004). Ice creams have been in existence for over 300 years. They are now relatively stable components of the dairy industry and have been manufactured commercially since the middle of the $19^{\text {th }}$ century (Davis et al., 2009). The development of these products in this thesis consisted of two stages with product development objectives in each stage. An overview of the development process for the ice creams is displayed in Figure 1.11. The first stage is concerned with culinary mechanisms at a molecular level. The mechanisms examined were food system formation and stability (objective 1) and flavour release (objective 2). Banana was selected as the primary ingredient in the food pairings as it is a pleasant fruit that is one of the most produced and consumed fruits throughout the world (Mayr et al., 2003; Jordán et al,. 2001). The results of the first stage were directly incorporated into the production of the novel ice cream products. The second stage is concerned with consumer preference. This involved exploring novel flavour selection (pairing foods with the banana) and consumer hedonic reactions (objective 3), and market research (objective 4).


Figure 1.12 Schematic overview of the development process for the novel ice creams.

## Chapter 1

1.9.3 Research aims and objectives

An overview of the objectives, questions and principal investigations involved in development of novel food products is listed in Table 1.3. The main aim of this project was to produce innovative food products based on molecular gastronomy principles.

The main objectives are to:

- Optimise the formation and stability of a food dispersion model system.
- Optimise the flavour volatile release of selective volatiles from the optimised dispersion model system.
- Investigate novel flavour combinations in terms of sensory hedonic responses and flavour volatile interactions.
- Produce innovative food products based on the findings of the previous objectives and assess their consumer acceptability and preference for these products and their suitability to a specific market sector.

Table 1.3 Objectives, questions and principal investigations involved in development of novel ice cream products.

| Objective 1: Optimise the formation and stability of a food dispersion model system. |  |
| :--- | :--- | :--- |
| Questions | Investigation |
| II. $\quad$What is the optimum emulsion formula for maximum <br> formation and physical stability of the oil in water emulsion? | Analysis of emulsion storage stability and use of response surface <br> methodology to model the effects of ingredients on emulsion storage <br> stability and establish optimum concentrations. |
| III. $\quad$What is the optimum emulsion formula for minimum mean <br> emulsion droplet diameter? | Image analysis of mean droplet diameter and use of response <br> surface methodology to model the effects of ingredients on mean <br> emulsion droplet diameter and establish optimum concentrations. |
| Objective 2: Optimise the flavour volatile release of selective volatiles from the optimised dispersion model system. |  |

Table 1.3 (continued) Objectives, questions and principal investigations involved in development of novel ice cream products.

| II. Do consumers consider the screened food pairings to be acceptable? | Preform a consumer sensory panel on the food pairings. |
| :---: | :---: |
| III. What are reasons for the consumer panel hedonic response to the food pairings? | Conduct volatile profile analysis and descriptive sensory analysis on the food pairings and correlate the results. |
| Objective 4: Produce innovative food products based on the findings of the previous objectives and assess their consumer acceptability and preference for these products and their suitability to a specific market sector. |  |
| Questions | Investigation |
| I. How can the bacon and basmati rice flavours be successfully transferred to the ice creams? | Carry out preliminary sensory analysis on various methods of flavour transfer from bacon and basmati rice. |
| II. What are the preferred flavour ratios of banana to bacon and of banana to basmati rice in the ice cream formulations? | Complete sensory analysis on the different formulations of banana and bacon ice cream and of banana and basmati rice ice cream to obtain those which are most preferred. |
| III. What are the pH and salt levels of the ice creams? Are they in line with the findings from objective 2 ? | Perform pH and salt determination analysis of the two ice cream samples. |
| $\begin{array}{ll} \hline \text { IV. What is the most suitable market sector for the two ice cream } \\ & \text { products and ultimately are they liked by consumer panels? } \end{array}$ | Conduct quantitative sensory analysis (affective tests) on the two ice cream products with two sensory panels representing different markets (general and specific). |
| V. What was the reason behind panellists' hedonic responses to the two ice cream products? | Gather qualitative data through sensory analysis of the two ice cream products. |

## Chapter 2

Development of novel ice cream products using molecular gastronomy theories

Optimisation of emulsion model system formation and stability

- section 2.2


Droplet size analysis


Stability analysis

Optimisation of flavour emission from the optimised emulsion - section 2.3


Volatile and sensory evaluation of novel food pairings - section 2.4


Development of novel flavoured ice creams - section 2.5


Figure 2.1 Overview of experimental plan.

### 2.1 Ethical considerations

Prior to commencing this study, it was a requirement that ethical clearance be granted from DIT Research Ethics Committee before conducting any sensory tests using members of the public. It is believed that sensory testing has the potential to cause severe illness and even death, e.g. due to food poisoning or the ingestion of toxic ingredients. Therefore, it was essential all aspects of testing were considered to ensure all procedures and practices conformed to all legal and ethical requirements. Declaration of research ethics and risk assessment documentation was sent to the DIT Research Ethics Committee, outlining procedures for the protection of sensory panellists, safety of sample ingredients, sample preparation and test protocol. The project was granted ethical clearance, subject to individuals being fully informed of the project and completing consent forms (appendix A).

### 2.2 Optimisation of emulsion formation and stability

### 2.2.1 Emulsion raw materials

Soy lecithin (Kelkin, Dublin, Ireland) was purchased in a local supermarket (Dunnes Stores, Dublin, Ireland). Xanthan gum was kindly donated by Chemcolloids Ltd. (Cheshire, UK). Sunflower oil (Basso Fedele and Figli, Avellino, Italy) was purchased from a local food supplier.

### 2.2.2 Experimental design

A Box Behnken experimental design was chosen, and 15 random order ( 2 centre point replicates) experimental settings were generated with 3 factors ( $x_{1}-x_{3}$ ), each with low $(-\alpha)$, medium $(\alpha)$ and high settings $(+\alpha)$, using $R 2.14 .1$ software package ( $R$ Development Team, Vienna, Austria) (Table 2.1). The effect of three independent coded variables on day 14 of storage, $x_{1}$ (sunflower oil concentration 10-20 \% v/v), $x_{2}$ (lecithin concentration $1-5 \% \mathrm{w} / \mathrm{v}$ ) and $x_{3}$ (xanthan gum concentration $0.01-0.3 \%$ $\mathrm{w} / \mathrm{v}$ ) on emulsion storage stability ratio (SR) was studied. This particular experimental design was selected as it proved to be the most economical experimental design (13 experimental conditions) for the evaluation of the multiple experimental parameters, while taking into account the interactions between variables and allowing for optimisation.

Table 2.1 Experimental design with level of factors (low ( $-\alpha$ ), medium ( $\alpha$ ) and high $(+\alpha)$ according to the Box Behnken design.

| Run | Sunflower oil (\% v/v) | Lecithin (\% w/v) | Xanthan gum (\% w/v) |
| :---: | :---: | :---: | :---: |
| 1 | 15 | 5 | 0.01 |
| 2 | 10 | 3 | 0.3 |
| 3 | 10 | 3 | 0.01 |
| 4 | 15 | 3 | 0.16 |
| 5 | 10 | 1 | 0.16 |
| 6 | 15 | 1 | 0.01 |
| 7 | 10 | 5 | 0.16 |
| 8 | 20 | 3 | 0.01 |
| 9 | 20 | 5 | 0.16 |
| 10 | 15 | 3 | 0.3 |
| $11^{\text {c }}$ | 15 | 3 | 0.16 |
| $12^{\text {c }}$ | 15 | 3 | 0.16 |
| 13 | 20 | 1 | 0.16 |
| 14 | 20 |  | 0.3 |
| 15 | 15 |  | 0.3 |

[^0]Selection of emulsion ingredients and their preliminary experimental ranges were based on literature research for sunflower oil (Mirhosseini et al., 2008b; Comas et al., 2006), lecithin (Klinkersorn \& McClemnets, 2009; Surh et al., 2008; Thakur et al., 2007; Malone et al., 2003) and xanthan gum (Mirhosseini et al., 2008a: Sun et al., 2007; Hemar et al., 2001). Furthermore, the preliminary experimental ranges were narrowed down to the experimental ranges based on preliminary experimentation where emulsion gravitational stability was measured.

Box Behnken designs are 3 levels factorial designs that have all points as centroids of the faces of a hypercube with dimensions equal to the number of factors (Box \& Behnken, 1960). Thus, all these treatments lay on a single hyper-sphere and are equally distant from the centre. Box Behnken designs are especially useful for experiments in 3 to 7 experimental factors (Schrevens \& Portier, 2006). Box Behnken experimental designs has previously been employed to investigate emulsion stability (Martinez et al., 2011).

For statistical analysis the variables were centred in the interval $[-1,1]$ by:
$\mathrm{x}_{\mathrm{n}}=\mathrm{x}-\mathrm{x}_{\text {min }} /\left(\left(\mathrm{x}_{\text {max }}-\mathrm{x}_{\text {min }}\right) / 2\right)$

Where x and $\mathrm{x}_{\mathrm{n}}$ are the original and the coded variable respectively and $\mathrm{x}_{\text {max }}$ and $\mathrm{x}_{\text {min }}$ are the maximum and minimum of the variable x . SR measurements were taken on day 14 , measurements were repeated three times, and experiments were conducted in triplicate, resulting in nine measurements in total for each experimental condition for SR. DS was measured at day $0,1,7$ and 14 , and experiments were conducted in triplicate. The optimal level of three independent variables ( $x_{1}, x_{2}$ and $x_{3}$ ) which led to the desirable multi response goals (minimise DS and maximise SR) was determined by
graphical and numerical optimisation procedures. Contour plots and canonical analysis were employed in order to deduce workable optimum conditions.

### 2.2.3 Preparation of emulsions

Surfactant solutions were prepared by dispersing the lecithin (1-5\% w/v) and xanthan gum ( $0.01-0.3 \% \mathrm{w} / \mathrm{v}$ ) in distilled water and mixing for 1.5 hours at $60^{\circ} \mathrm{C}$ (Sznitowska et al., 2002) using a magnetic stirring bar and magnetic stirrer hotplate (Stuart CB162, Bibby-scientific, Staffordshire, UK). Sunflower oil (10-20 \% v/v) was then heated to $60^{\circ} \mathrm{C}$ and slowly added to the aqueous phase while stirring. This preemulsion was allowed to mix for a further 5 minutes before being homogenised with a high shear blender (Silverson L4R, Silverson Machines Ltd., Chesham, UK) for 3 minutes at $8,000 \mathrm{rpm}$. The resulting emulsion was allowed to cool to room temperature. Thirty mL of emulsion sample was transferred to a graduated 40 mL plastic test tube ( $84 \times 30 \mathrm{~mm}$ polypropylene with screw cap, Sarstedt Ltd., Wexford, Ireland), tightly sealed with a plastic cap, and stored in a dark incubator (Innova 42, New Brunswick Scientific, Enfield, CT, USA) for 14 days at ambient room temperature $\left(23{ }^{\circ} \mathrm{C}+/-1^{\circ} \mathrm{C}\right)$.

### 2.2.4 Analysis of emulsion stability

Emulsion stability ratio (SR) was calculated as a percentage of the initial emulsion height in the tube (HE), height of cream layer (HC) and height of sedimentation phase (HS) (Mirhosseini et al., 2008b):

$$
S R=\frac{(H E-(H C+H S))}{H E} \times 100
$$

Equation 2.2

Emulsion stability measurements were taken on day 14, experimental determinations were carried out in triplicate and each measurement was recorded three times resulting in nine measurements in total for each sample. Images of emulsions during storage are presented in Figure 2.2.


Figure 2.2 Images of an emulsion with (a) $100 \%$ SR after 14 days of storage and (b) an emulsion with phase separation after 14 days of storage.

### 2.2.5 Analysis of mean droplet diameter by image analysis

Image acquisition was conducted using an optical microscope (Olympus BH2, Olympus, Southend-on-Sea, UK) with a 400 magnification coupled with a digital camera (Canon DSLR EOS D30, Canon Ireland, Dublin, Ireland). The images were then transferred to a personal computer, and were processed following the method sequence of other studies involving binarization and droplet quantification (Silva et al., 2010; Trindade et al., 2008; Freire et al., 2005). The image processing procedure was developed using an automated program developed in MATLAB 7.0 (The Math Works, Inc., Natick, MA, USA). The Image J software (Version 1.451, U.S National Institute of Health, Bethesda, MD, USA) and a stage micrometer (OB-M stage micrometer,

Olympus Imaging and Audio Ltd., Essex, UK) of known size were used for calibration of the droplet diameter.

Before analysis each sample was diluted with distilled water in a ratio of 1:1000. A small drop of the diluted emulsion (approximately 0.1 ml ) was transferred to a microscope slide and viewed at 400 magnification on the optical microscope with the digital camera for imaging capturing. Measurements were taken on day $0,1,7$ and 14 . On each day of analysis, an average of 23 droplets per image and an average of 22 different images per sample were analysed providing an average of 500 droplets for each sample and an average of 1,500 droplets for each condition per point. At the end of the experiment over 4,000 images were analysed in total. An example of an intensity image and its corresponding binary image is presented in Figure 2.3.


Figure 2.3 Microscopic images of (a) intensity image and (b) binary image of an oil in water emulsion.

The bar corresponds to $20 \mu \mathrm{~m}$.

### 2.2.6 Statistical analysis of emulsion formation and stability data

### 2.2.6.1 Regression modelling

All statistical analysis was carried out using R software (Version 2.14.1, R Development Team, Vienna, Austria). The effects of three independent variables (sunflower oil concentration $\left(x_{1}\right)$, lecithin concentration $\left(x_{2}\right)$ and xanthan gum concentration $\left(x_{3}\right)$ ) on the emulsion stability of an oil in water emulsion and their interaction were analysed using polynomial regression analysis. A second order polynomial equation for the three dependent variables $\left(x_{1}-x_{3}\right)$ was established to fit the experimental data;

$$
\begin{aligned}
& \mathrm{SR}=\beta_{0}+\beta_{1} \mathrm{x}_{1}+\beta_{2} \mathrm{x}_{2}+\beta_{3} \mathrm{x}_{3}+\beta_{12} \mathrm{x}_{1} \mathrm{x}_{2}+\beta_{13} \mathrm{x}_{1} \mathrm{x}_{3}+\beta_{23} \mathrm{x}_{2} \mathrm{x}_{3}+ \\
& \beta_{11} \mathrm{x}_{1}^{2}+\beta_{22} \mathrm{x}_{2}^{2}+\beta_{33} \mathrm{x}_{3}^{2}+\varepsilon \\
& \text { Equation } 2.3
\end{aligned}
$$

Where $S R$ is the emulsion stability ratio response, $x_{1} \ldots . x_{1}{ }^{2} \ldots . x_{1} x_{2} \ldots$ are the independent linear, quadratic and interaction terms. In the model, $\beta_{0}$ is the intercept term, $\beta_{1 \ldots .} \beta_{11 \ldots .} \beta_{12 \ldots}$.are the regression coefficients for the linear, quadratic and interaction effect terms and $\varepsilon$ is an error term corresponding to an independent normal distribution. Estimation of the regression parameters and their standard errors was done using the least-square technique (Myers et al., 2009). The effects of the independent variables on the mean droplet diameter (DS) of an oil in water emulsion and their interaction were analysed using a primary model to describe the dependence of the DS on storage time and a secondary model to describe the dependence of the primary model parameters on the design variables (Aguirre et al., 2009).

The primary model chosen to best describe the kinetics of DS during storage after graphical analysis was an apparent first order kinetic model described by;

$$
\mathrm{DS}=\text { Asym }+\left(\mathrm{R}_{0}+\left(\mathrm{R}_{0}-\text { Asym }\right)\right) \mathrm{e}^{-\mathrm{e}^{\operatorname{lr}} \text { time }}+\varepsilon \quad \text { Equation } 2.4
$$

Where Asym, $R_{0}$ and lrc are the asymptotic DS at long storage time, $R_{0}$ is the initial droplet diameter, Asym is the final droplet diameter, lrc is the natural logarithm transformation of the apparent first order rate constant $\left(\mathrm{Day}^{-1}\right)$ and $\varepsilon$ is an error term corresponding. The secondary modelling of the three parameters Asym, $R_{0}$ and $l r c$ was done using a second order polynomial dependence on the coded variables $x_{1}, x_{2}$ and $x_{3}$. The building procedure to obtain a model with a minimum set of dependence of the three parameters with the experimental variables followed these steps:

1. An initial model was built using a second order polynomial dependence of the parameters $A s y m, R_{0}$ and $l r c$ with the normalised variables $x_{1}, x_{2}$ and $x_{3}$.
2. A summary of the model was produced with $t$-statistics for each individual model coefficient.
3. Based on the $t$-test statistics of significance, non-significant terms of the model were eliminated (Pinheiro \& Bates, 2000) and a new model was built.
4. A summary of the new model was produced with new $t$-statistics for each model term.
5. A logarithm likelihood ratio test (Bates \& Watts, 1988) was employed to compare the new model with the previous one and decide if the simplification obtained was statistically significant or not in terms of capacity to describe the data (i.e. the log-likelihood).
6. Steps 1-5 were repeated with new simplifications proposed until a satisfactory model reduction was achieved.

The adequacy of the model was determined using graphical analysis and examining the coefficient of determination $\left(R^{2}\right)$ (Lee et al., 2000). It is suggested that $R^{2}$
should be at least 0.80 for a good global fit of a model (Joglekar \& May, 1987). Analysis was performed in triplicate and data was reported as the estimated parameter $\pm$ standard error (SE). To determine the significance of a model parameter, the student's $t$ test was used. Differences were considered to be statistically significant at $\mathrm{p} \leq 0.05$. The RSM libraries (Lenth, 2009) and nls (nonlinear least squares) from the R software (Version 2.14.1, R Development Team, Vienna, Austria) were used.

### 2.2.6.2 Multicriteria optimisation

The optimal conditions for the targeted responses were generated by R 2.14 .1 software package. The NSGA II MOEA numeric optimisation algorithm from the R library MCO (Multi Criterion Optimisation) (Trautmann et al., 2010) was employed to study the simultaneous optimisation of SR and DS. A Pareto front from the multiple response optimisations was defined to maximise the SR and minimise the DS within the experimental range studied.

### 2.2.7 Validation of optimal conditions for emulsion stability and droplet diameter

The adequacy of response surface models for predicting optimum response values was verified by conducting experiments under one of the sets of optimum conditions devised in the Pareto front (sunflower oil ( $\mathrm{x}_{1}$ ) $19.02 \%$, lecithin ( $\mathrm{x}_{2}$ ) $1.2 \%$, xanthan gum ( $\mathrm{x}_{3}$ ) $0.28 \%$ ). The experimental predicted values of the responses were compared in order to check the validity of the models.

### 2.3 Optimisation of flavour emission from the emulsion

### 2.3.1 Flavoured emulsion raw materials

Sunflower oil, soy lecithin and xanthan gum were sourced as outlined in section 2.2.1. Isoamyl acetate, furfuryl acetate, citric acid, hydrogen sodium carbonate, SPME fibre, 20 mL glass vial and Teflon coated rubber septa and aluminium screw caps were purchased from Sigma Aldrich (Sigma Aldrich Ireland Ltd., Dublin, Ireland).

### 2.3.2 Experimental design

A Central Composite Faced Centred experimental design (CCF) was employed to (1) study the main effect and combined effect of these independent variables on the volatile emission of isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ and furfuryl acetate $\left(\mathrm{Y}_{2}\right)$ from the oil in water emulsion, (2) create empirical models between the variables and (3) optimise the physicochemical conditions for maximum volatile emission. The effect of four independent variables namely, $x_{1}$ (isoamyl acetate concentration 50-90 ppm), $x_{2}$ (furfuryl acetate concentration 20-30 ppm), $x_{3}(\mathrm{pH} 5-7)$ and $x_{4}$ (salt concentration 0.1 $-2 \%$ ) on the volatile emission from an oil in water emulsion were studied using RSM.

Experimental ranges for isoamyl acetate and furfuryl acetate were selected based on the Flavour and Extract Manufacturers Association (FEMA) reported uses in literature (Burdock, 2010). In addition, preliminary sensory analysis ( $\mathrm{n}=12$ ) was conducted, during which panellists successfully ranked emulsions containing various concentrations of isoamyl acetate and furfuryl acetate in order of concentration. Experimental ranges for salt and pH were selected bases on typical salt and pH levels in food products.

Central Composite Designs (CCD) are experimental designs that are composed of a Factorial Design (the points at the vertices of the cube) and by a Star Design which allow for estimation of curvature (Leardi, 2009). The star points represent new extreme values (low and high) for each factor in the design (Schrevens \& De Rijck, 1999). CCF is a subset of CCD in which the location of the star points corresponds to the centre of the faces of the cube and 3 levels of each factor are required (Leardi, 2009). CCD designs have been employed by previous authors for the investigation of ingredient interactions on the headspace release of volatile compounds from emulsion model systems (Mirhosseini \& Tan, 2009; Mirhosseini et al., 2008c; Mirhosseini et al., 2008d).

A CCF with 27 random order settings (3 centre point replicates) was chosen using Modde 5.0 software (Umetrics UK, Cheshire, UK) as this particular experimental design proved to be the most economical design for this particular experimental work. The volatile release of each flavour compound was expressed by the peak area recorded by using GC/MS. The concentration of each volatile in the headspace above the emulsion was considered as response variables. This was calculated by comparing the response for both of the volatiles to that of an aqueous standard calibration curve. The response values were calculated as the concentration (in ppm) in the headspace of the emulsions (calculated based on a standard curve constructed using an aqueous solution of isoamyl acetate and furfuryl acetate) divided by the concentration of the corresponding aqueous solution.

### 2.3.3 Preparation of flavoured emulsions

Emulsions were prepared as described in section 2.2 .3 with the following modifications: surfactant solutions were prepared by dispersing the lecithin and xanthan gum in distilled water and mixing for 1.5 hours at $60^{\circ} \mathrm{C}$ using a magnetic stirring bar and magnetic stirrer hotplate (Stuart CB162, Bibby-scientific, Staffordshire, UK). The lipid phase was then heated to $60^{\circ} \mathrm{C}$ and the flavour compounds were then added by micro pipette. The lipid phase was then slowly added to the aqueous phase while stirring. This pre-emulsion was allowed to mix for a further 5 minutes before being homogenised with a high shear blender (Silverson L4R, Silverson Machines Ltd., Chesham, UK) for 3 minutes at $8,000 \mathrm{rpm} . \mathrm{NaCl}$ was then added to the emulsion and allowed to dissolve under gentle stirring for 1-2 minutes. The pH was adjusted within the range of pH 5-7 using a pH meter (Thermo scientific, Orion 2 Star, Essex, UK) with 0.1 M citric acid and 0.5 M sodium hydrogen carbonate.

### 2.3.4 Analysis of volatile emission from the emulsion

### 2.3.4.1 Emulsion headspace volatile analysis

A solid phase microextraction (SPME) device (Supelco, Bellefonte, PA, USA) with a pre-conditioned 10 mm SPME fibre coated with $65 \mu \mathrm{~m}$ PolydimethylsiloxaneDivinylbenzene (PDMS/DVB) was used for the extraction of the headspace volatiles. Ten mL aliquots of the emulsion were transferred into 20 mL headspace glass vials (O.D $2.25 \mathrm{~mm} \times \mathrm{H} 75 \mathrm{~mm}$ ) which were sealed with a stainless steel magnetic screw cap fitted with a Polytetrafluoroethylene (PTFE)/silicone septum (septum thickness 1.3 $\mathrm{mm})$. A PTFE coated micro stirring bar ( $13 \mathrm{~mm} \times 8 \mathrm{~mm}$ ) was used to simulate shear stress in the mouth. A shear rate of approximately $150 \mathrm{~s}^{-1}$ was achieved by stirring at

450 rpm (Rabe et al., 2003). The vials were placed in a water bath set at $40^{\circ} \mathrm{C}$ for a preequilibrium time of 5 minutes and the SPME fibre was exposed to the headspace of the samples for an extraction time of 30 minutes. To determine optimal extraction time, the SPME fibre was exposed to the headspace above the emulsions containing different concentrations of the flavour compounds for various lengths of time (5-60 minutes) at $40^{\circ} \mathrm{C}$. Results from this preliminary experiment indicated that an extraction time of 30 minutes was sufficient to facilitate equilibration. At these concentrations, the volatiles were deemed to be at equilibrium and extending sample time beyond this point was unwarranted.

### 2.3.4.2 Gas chromatography mass spectrometry (GC/MS) conditions

Analysis of the volatile compounds absorbed on the fibre was carried out using a Varian 3800 GC coupled to a Varian Saturn 2000 ion trap mass spectrometer (Varian Chromatography Systems, Walnut Creek, CA, USA). Separation of the volatiles was accomplished on a ZB-wax column (ZB-5MS- $15 \mathrm{~m} \times 0.25 \mathrm{~mm}$ i.d., $0.25 \mu \mathrm{~m}$ film, Torrance, CA, USA). Helium, at a flow rate of $1 \mathrm{~mL} /$ minute was used as the carrier gas. Thermal desorption of the compounds took place in the GC injection port (1079 Programmable Temperature Vaporising (PTV) injector), equipped with a 0.75 mm i.d. splitless glass linear, at $250^{\circ} \mathrm{C}$. The split valve was initially closed, after 5 minutes it was opened (ratio 1:100) for the remainder of the run. The SPME fibre was removed from the headspace of the samples and thermally desorbed at $250{ }^{\circ} \mathrm{C}$ in the GC injection port for 3 minutes. The fibre was cleaned after every two extractions to ensure no carryover between samples. The stirrer bar was cleaned after every extraction by soaking it in acetone for at least 15 minutes and then rinsing with $40 \%$ ethanol before re-use (LayKeow, 1998).

Oven temperature was programmed at $40^{\circ} \mathrm{C}$ for 1.5 minutes, then ramped to 80 ${ }^{\circ} \mathrm{C}$ at $4^{\circ} \mathrm{C} /$ minute and finally raised to $250^{\circ} \mathrm{C}$ at $20^{\circ} \mathrm{C} /$ minute and held for 5 minutes. The MS transfer line temperature was set to $260^{\circ} \mathrm{C}$. The mass spectrometer was tuned using autotune procedures and masses from $\mathrm{m} / \mathrm{z} 40$ to 180 were recorded after electron impact ionization (EI) under EI auto mode. Isoamyl acetate and furfuryl acetate were initially identified using a Varian 450-GC equipped with a Varian 320-MS triple quadrupole (Varian Chromatography Systems, Walnut Creek, CA, USA) and CombiPal Autosampler (CTC Analytics, Zwingen, Switzerland). Compounds were also identified by use of authenticated standards and by matching mass spectra with the data stored in the National Institute of Standards and Technology (NIST) library of standard compounds. The data reported was the mean of three extraction replicates for each individual peak in the total ion chromatogram (TIC). Compounds were quantified by reference to external calibration curves constructed using the same authenticated standards. The response values were calculated as the concentration (in parts per million $(\mathrm{ppm})$ ) in the headspace of the emulsions (calculated based on a standard curve constructed using an aqueous solution of isoamyl acetate and furfuryl acetate) divided by the concentration of the corresponding aqueous solution. The standard curves had $R^{2}$ values of 0.996 and 0.976 for the isoamyl acetate and furfuryl acetate respectively.

### 2.3.5 Statistical analysis of volatile emission data

All statistical analysis was carried out using Modde 5.0 (MKS Instruments UK Ltd., Umetrics UK, Cheshire, UK) software. The effects of the independent variables on the volatile flavour release of isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ and furfuryl acetate $\left(\mathrm{Y}_{2}\right)$ from an oil in water emulsion and their interaction were analysed using polynomial regression
analysis and analysis of variance (ANOVA). A second order polynomial equation for dependent variables was established to fit the experimental data.

The proposed generalised second order polynomial equation is given as;

$$
\begin{aligned}
& \mathrm{SR}=\beta_{0}+\beta_{1} \mathrm{x}_{1}+\beta_{2} \mathrm{x}_{2}+\beta_{3} \mathrm{x}_{3}+\beta_{4} \mathrm{x}_{4}+\beta_{12} \mathrm{x}_{1} \mathrm{x}_{2}+\beta_{13} \mathrm{x}_{1} \mathrm{x}_{3}+\beta_{14} \mathrm{x}_{1} \mathrm{x}_{4}+ \\
& \beta_{23} \mathrm{x}_{2} \mathrm{x}_{3}+\beta_{24} \mathrm{x}_{2} \mathrm{x}_{4}+\beta_{34} \mathrm{x}_{3} \mathrm{x}_{4}+\beta_{11} \mathrm{x}_{1}^{2}+\beta_{22} \mathrm{x}_{2}^{2}+\beta_{33} \mathrm{x}_{3}^{2}+\beta_{44} \mathrm{x}_{4}^{2}+\varepsilon
\end{aligned}
$$

Equation 2.5

Where Yi is the predicted response, $\mathrm{x}_{1} \ldots . \mathrm{x}_{1}{ }^{2} \ldots . \mathrm{x}_{1} \mathrm{x}_{2} \ldots$. are the independent linear, quadratic and interaction terms. In the model, $\beta_{0}$ is the intercept term, $\beta_{1} \ldots$ $\beta_{11 \ldots .} \beta_{12} \ldots$.are the regression coefficients for the linear, quadratic and interaction effect terms and $\varepsilon$ is an error term corresponding to an independent normal distribution. Determination of the different multiple regression parameters was achieved by employing the least-square technique (Myers et al., 2009). The adequacy of the model was determined using graphical analysis, lack of fit test and coefficient of determination ${ }^{\left(R^{2}\right)}$ (Lee et al., 2000). Analysis was performed in triplicate and data was reported as the mean $\pm$ standard error (SE). To determine the significance of a model parameter, the student $t$-test was used. Differences were considered to be statistically significant at $\mathrm{p} \leq$ 0.05 .

### 2.3.6 Validation of optimal conditions for headspace volatile emission

The adequacy of response surface models for predicting the optimum response values was verified by conducting experiments under the recommended optimum conditions devised in the optimiser function of the Modde 5.0 software (isoamyl acetate $90 \mathrm{ppm}\left(\mathrm{x}_{1}\right)$, furfuryl acetate $30 \mathrm{ppm}\left(\mathrm{x}_{2}\right)$, salt $2 \% \mathrm{w} / \mathrm{v}\left(\mathrm{x}_{3}\right)$ and $\left.\mathrm{pH} 6.02\left(\mathrm{x}_{4}\right)\right)$. The
experimental predicted values of the responses were compared in order to check the validity of the models.

### 2.4 Volatile and sensory evaluation of novel food pairings

The volatile and sensory evaluation of novel food pairings is summarised in Figure 2.4.


Figure 2.4 Schematic overview of procedures used in for the volatile and sensory evaluation of novel food pairings.

### 2.4.1 Food pairing raw materials

Gruyère cheese (Simply Better, Dunnes Stores Ltd., Dublin, Ireland), blue cheese (Farmhouse cheesemakers, Tipperary, Ireland), extra virgin olive oil (Don Carlos, Hacienda Don Carlos, Sevilla, Spain), soy sauce (Kikkoman, Kikkoman Trading Europe GmbH, Dusseldolf, Germany), whole grain mustard (Edmond Fallot, Beaune, France), fresh mackerel (Atlantic Mackerel (Scomber scombrus)), bacon (St. Bernard, mild cured back rashers, Dunnes Stores Ltd., Dublin, Ireland), basmati rice (Dunnes Basmati Rice, Dunnes Stores Ltd., Dublin, Ireland) and bananas (Cavendish, origin Costa Rica) (Fyffe plc, Dublin, Ireland) were purchased from the local supermarket (Dunnes Stores, Dublin, Ireland). Bananas were purchased as close to full ripeness as possible (yellow with green tips \& necks, stage 5), and allowed to fully ripen based on a commercial peel colour scale (entirely yellow, stage 6) (Figure 2.5). All food ingredients were purchased on the day of analysis.


Figure 2.5 Commercial banana colour guide.
Source: adapted from Chiquita Brands International Inc., Charlotte, NC, USA.

### 2.4.2 Screening of food pairings by focus groups

### 2.4.2.1 Selection of food pairings

Selection of the food pairing samples was performed through preliminary research and experimentation (focus groups and sensory panels). This preliminary work involved selecting eight food pairings with banana (bacon, basmati rice, extra virgin olive oil, blue cheese, Gruyère cheese, mackerel, soy sauce and whole grain mustard) which were generated using the online software package provide on Foodpairing.com (Sense for Taste, Bruges, Belgium). The food pairing software suggested over 40 foods to be paired with banana. In order to reduce this number to a more manageable quantity (for experimental purposes), factors such as the availability and expense of the foods were taken into account. Furthermore, the foods were ranked based on the quantity of suggested pairings each food had with one another (in addition to their pairings with banana). The eight foods with the highest quantity of food pairings were subsequently selected. For example, it was found that bacon was paired with 14 other foods, while tomato only had 10 pairings, thus tomato was subsequently not selected for further analysis. These eight food pairings were then subjected to further screening tests to reduce the sample number to allow for sensory and instrumental work (focus groups and sensory evaluation).

### 2.4.2.2 Conducting the focus groups

Three separate focus groups ( $\mathrm{n}=19$ in total) were conducted, during which information regarding the participant's attitudes and opinions towards the pairing of novel foods were explored. Participants involved were students from the School of Culinary Arts and Food Technology and from the School of Food Science and

Environmental Health, Dublin Institute of Technology, Dublin. The focus groups were conducted according to standard procedures (Krueger \& Casey, 2009; Morgan, 1998; Morgan \& Scannell, 1998). The setting was a conference room, and audio recordings of the focus groups were conducted on personal computers using sound recording and editing software (Audacity 1.3, The Audacity Development Team). Each focus group lasted between 90-120 minutes and was guided by a moderator. Participants were provided with information regarding the general rules for performing each session, and were familiarised with the subject matter "food pairings" via a short presentation performed by the moderator. A topic guide was developed prior to the field work and consisted of open discussions on: (1) the concept of pairing foods based on flavours, (2) traditional/known food pairings that are successful and (3) novel food pairings. Visual cues (pictures of food pairings) were used throughout the focus groups sessions to relate to the food pairings being discussed. Following this participants were asked to evaluate the orthonasal aroma of the food pairings (section 2.4.2.4). A final open discussion regarding the food pairing samples that were evaluated took place following the sensory evaluation session.

### 2.4.2.3 Preparation of samples for orthonasal sensory evaluation

Prior to conducting the focus groups, the bacon was cooked in an oven (Rational Combi Dämpfer, Rational UK, Luton, UK) at $190^{\circ} \mathrm{C}$ for 9 minutes and then turned and cooked for a further 9 minutes. Basmati rice was cooked in water at a ratio of 1.5:1 water: rice (Limpawattana et al., 2008) until deemed cooked (tender with a little bite). The mackerel was baked in an oven (Rational Combi Dämpfer, Rational UK, Luton, UK) at $190^{\circ} \mathrm{C}$ for approximately 8 minutes until the centre of the fish reached $\geq 70^{\circ} \mathrm{C}$ for over 2 minutes (Cox \& Abu-Ghannam, 2013). Once the bacon, basmati rice and
mackerel were cooked, they were allowed to cool to below $5{ }^{\circ} \mathrm{C}$. Ten grams (g) of Gruyère cheese, blue cheese, cooked basmati rice, olive oil, soy sauce, 5 g of mustard, 4 g of bacon and 2.5 g of cooked mackerel were place separately in the individual sample containers with lids on and were then stored below $5{ }^{\circ} \mathrm{C}$ until the sensory evaluation took place (approximately one hour). Sample quantities were selected based on preliminary testing, where a perceived aroma balance was achieved through preliminary sensory analysis $(\mathrm{n}=5)$. To reduce oxidation of the bananas, they were prepared by slicing only as required, 5 g of banana was sliced up and placed in the sample containers.

### 2.4.2.4 Orthonasal sensory evaluation of food pairings

The orthonasal aroma of the food pairings was evaluated through sensory analysis according to ISO 5496:2006 (ISO, 2006). Sensory analysis was carried out in a sensory laboratory under guidelines and conditions according to ISO 8589:2010 (ISO, 2010). The panellists worked in a single booth under defined conditions of $22{ }^{\circ} \mathrm{C}$ and white light. Samples prepared at least 30 minutes before the test to allow time for vapour pressure to reach equilibrium at ambient temperature. The food pairings were placed in three-digit random number coded opaque plastic containers with opaque lids so that respondents could smell them without seeing. Five minutes before the presentation of the food, small holes were made with a needle, through which the aromas were released (Raz et al., 2008). Panellists were presented with samples of banana to which they retained for the entire evaluation session. The 8 samples to be paired with the banana were presented in a monadic sequential randomised order. Panellists were instructed to evaluate the banana sample and the paired sample together
(three short sniffs with the mouth closed). Panellists evaluated the orthonasal aroma acceptability of the food pairings on a 9 point hedonic scale, where $9=$ "like extremely", $5=$ "neither like nor dislike" and $1=$ "dislike extremely" (see appendix B).

### 2.4.3 Consumer sensory evaluation of food pairings

From the results of this preliminary sensory work, it was found that the banana food pairings of basmati rice $(B+R)$, bacon $(B+B N)$ and extra virgin olive oil $(B+O)$ scored the highest mean acceptability scores for orthonasal aroma (see section 5.2.2). Hence, these samples were used for the consumer sensory panel, descriptive sensory analysis and organic volatile compound profiling. Untrained panellists ( $\mathrm{n}=85$ ) were asked to evaluate the taste of the three food pairings with banana. The consumer panel consisted of staff and students from the School of Culinary Arts and Food Technology and from the School of Food Science and Environmental Health, Dublin Institute of Technology, Dublin. Sensory analysis was carried out in a sensory laboratory under guidelines and conditions according to ISO 8589:2010 (ISO, 2010). The panellists worked in a single booth under defined conditions of $22{ }^{\circ} \mathrm{C}$ and white light.

Prior to conducting the consumer panel, bacon and basmati rice were prepared as described in section 2.4.2.3. Sample quantity ratios were selected based on preliminary sensory testing ( $n=5$ ), where a perceived flavour balance was achieved. Cooked bacon and cooked rice were heated separately in a microwave oven (Sharp Plutonium Collection FR957, Cheshire, UK) at 600 W to above $63{ }^{\circ} \mathrm{C}$, and were held above this temperature in a bain maire until required (less than five min). To reduce oxidation of the bananas, they were prepared by slicing freshly as required. Twenty grams of both cooked bacon and cooked rice were served separately with 20 g of sliced
banana in the sample containers. Twenty grams of sliced banana were served in the sample containers with 10 ml of the olive oil. Samples were coded with three-digit random numbers and were presented in a monadic sequential randomised order to the panel. Panellists were instructed to consume unsalted crackers and mineral water to rinse their mouths between evaluating each sample (Bárcenas et al., 2001).

For the hedonic tests, consumers were instructed to evaluate taste acceptability of the paired samples on a 9 point hedonic scale, where $9=$ "like extremely", $5=$ "neither like nor dislike" and $1=$ "dislike extremely" (see appendix B). Additionally, the panellists were also asked to rank the samples in order of preference, from most preferred to least preferred (see appendix B). Panellists were encouraged to write comments regarding their opinion of the food pairings, these comments were coded (both positive and negative comments).

### 2.4.4 Descriptive sensory analysis (Free Choice Profiling)

Assessors ( $\mathrm{n}=28,11$ males, 17 females, aged 20-40) were recruited for orthonasal and retronasal aroma evaluation of seven food samples (banana (B), bacon $(B N)$, rice $(R)$, extra virgin olive oil $(O)$, banana and rice $(B+R)$ and banana and bacon $(\mathrm{B}+\mathrm{BN})$, and banana and extra virgin olive oil $(\mathrm{B}+\mathrm{O}))$ using Free Choice Profiling (FCP). Assessors were recruited from staff and students from the School of Culinary Arts, Food Technology and from the School of Food Science and Environmental Health, Dublin Institute of Technology, Dublin, and from the School of Food and Nutritional Sciences, University College Cork, Cork. All assessors had previous experience in sensory evaluation. The assessors were separated into three groups of 10 , 10 and 8 , a total of two sessions were carried out with each group; the first session was
a training session and the second session was a data collection session. Each session took place on separate days and took approximately 60-90 minutes to complete.

In the training session the assessors got a brief introduction into the procedure of FCP, during which descriptive lexicons were developed and defined for each sample with the help of some previously developed descriptive lexicons from the literature (see appendix C) (Delgado \& Guinard, 2011; Sekhon, et al., 2010; Limpawattana et al., 2008; Kanavouras et al., 2005; Timón et al., 2004; Jordán et al., 2001; Maw et al., 2001; Angerosa et al., 2000; Civille \& Lyons, 1996; Jeremiah et al., 1996; Morales et al., 1995). Basmati rice and bacon samples were prepared as described in section 2.4.2.3. The paired samples (banana and basmati rice $(B+R)$, banana and bacon $(B+B N)$ and banana and extra virgin olive oil $(\mathrm{B}+\mathrm{O})$ ) were served to assessors as described in section 2.4.3.

For the unpaired samples (banana, basmati rice, bacon and extra virgin olive oil); 20 g of heated cooked basmati rice, 20 g of heated cooked bacon, 10 ml of extra virgin olive oil and 20 g of sliced banana were placed separately in sample containers. All seven samples were coded with three-digit random numbers and were presented in a monadic sequential order during each session. Breaks between each sample evaluation were provided, during which assessors were instructed to consume unsalted crackers and mineral water to rinse their mouths.

Assessors were asked to evaluate the orthonasal aroma, followed by the retronasal aromas of the samples, and describe their sensory perception in their own words with the help of the descriptors provided. Assessors were asked to evaluate the intensity of the sensory attributes of the samples using a 15 cm line scale, anchored with the terms "weak/mild" and "strong" (Appendix C). Based on the individual descriptors,
personalised score sheets were created, and a consensual description list was generated from each panellist for each sample. At the beginning of the second session the assessors were asked to take a moment to read the consensual description list and to update their own list if desired. The assessors then proceeded to evaluate the intensity of the sample's attributes as described in the first session.

### 2.4.5 Organic volatile compound profile analysis

### 2.4.5.1 Headspace volatile analysis for volatile profiling of food pairings

Sample introduction was accomplished using a CTC Analytics Combipal Autosampler (Agilent, Cork, Ireland). A single 1 cm SPME device (Supelco, Bellefonte, PA, USA) with a 10 mm fibre coated with a $50 / 30 \mu \mathrm{~m}$ StableFlex Divinylbenzene/CarboxeNPolydimethylsiloxane (DVB/CAR/PDMS) fibre (Sulpelco, Bellafonte, PA, USA) was used for the extraction of the headspace volatiles. Sample preparation was as described in section 2.4.2.3. Initially, unpaired foods (banana, basmati rice, bacon and extra virgin olive oil) were analysed by HS-SPME GC/MS to assess the range of volatile compounds present in each sample and the response detected by the MS. Also this allowed for the determination of the quantities to be used during extraction of the food mixtures to ensure that the response of each individual food was within the same range. The quantities used for extraction of the individual foods were as follows: 5 g of bacon, 1 g of banana, 5 g of rice or 1 g of oil. These were weighed individually into 20 ml headspace SPME glass vials (O.D $2.25 \mathrm{~mm} \times \mathrm{H} 75 \mathrm{~mm}$ (Apex Scientific Ltd., Maynooth, Kildare, Ireland) which were sealed with a stainless steel magnetic screw cap fitted with a Polytetrafluoroethylene (PTFE)/silicone septum (septum thickness 1.3 mm ).

For evaluation of the paired samples (banana and basmati rice $(B+R)$, banana and bacon ( $\mathrm{B}+\mathrm{BN}$ ) and banana and extra virgin olive oil $(\mathrm{B}+\mathrm{O})$ ) the quantities used for extraction were as follows; banana 1 g : basmati rice $4 \mathrm{~g}(\mathrm{~B}+\mathrm{R})$, banana 1 g : bacon 4 g $(\mathrm{B}+\mathrm{BN})$ and banana 1 g : extra virgin olive oil $1 \mathrm{~g}(\mathrm{~B}+\mathrm{O})$. The choice of the ratios used for the paired samples analysis were established based on the preliminary volatile analysis of the individual food samples as discussed above. To determine optimal extraction time, the SPME fibre was exposed to the headspace above the samples for various lengths of time (5-60 minutes) at $40{ }^{\circ} \mathrm{C}$. Results from these preliminary experiments indicated that an extraction time of 20 minutes was sufficient to facilitate equilibration, and extending extraction time beyond this point was unwarranted. Sample mixtures were weighed into the vials and were placed in an incubation shaker chamber at $40{ }^{\circ} \mathrm{C}$ with pulsed agitation of 4 s at a shear rate of $150 \mathrm{~s}^{-1}$. Following a preequilibration step at $40{ }^{\circ} \mathrm{C}$ for 10 minutes the volatile compounds present in the headspace were extracted for 20 minutes.

### 2.4.5.2 GC/MS conditions for volatile profiling of food pairings

Analysis of the organic volatile compounds absorbed onto the fibre was carried out using a Varian 450 Gas Chromatograph (Varian Analytical Instruments, Harbour City, CA, USA) with temperature programmable split/splitless injector, oven cryogenics, equipped with a Varian 320 triple quadrupole Mass Spectrometer (Varian Chromatography Systems, Walnut Creek, CA, USA). Separation of the volatiles was accomplished on an Elite 5 MS column ( $60 \mathrm{~m} \mathrm{X} 0.25 \mu \mathrm{~m}, 0.25 \mu \mathrm{~m}$ film thickness, Perkin Elmar, MA, USA). Helium, at a flow rate of $1.2 \mathrm{~mL} /$ minute was used as the carrier gas. Thermal desorption of the compounds took place in the GC injection port at
$250{ }^{\circ} \mathrm{C}$ in splitless mode. The fibre remained in the injection port for 2 min . Oven temperature was programmed at $-60^{\circ} \mathrm{C}$ for 2 minutes, then ramped to $20^{\circ} \mathrm{C}$ at 50 ${ }^{\circ} \mathrm{C} /$ minute, then to $110^{\circ} \mathrm{C}$ at $4^{\circ} \mathrm{C} /$ minute, and finally raised to $250^{\circ} \mathrm{C}$ at $25^{\circ} \mathrm{C} /$ minute and held for 7 minutes. The transfer line temperature was set to $260^{\circ} \mathrm{C}$. The mass spectrometer was tuned using autotune procedures and operated in the scan mode within a mass range of $\mathrm{m} / \mathrm{z} 29$ to 350 at 2.5 scans/s. Ionisation was performed by electron impact at 70 eV . Peak areas were analysed and quantified using the Varian Star MS chromatography workstation software, version 6.9 .2 (Varian Analytical Instruments, Harbour City, CA, USA). Individual compounds were assigned quantification and qualifier ions to ensure that only the individual compounds were identified and quantified. Individual compounds were identified using mass spectral comparisons to the NIST 2005 mass spectral library. Quantification was performed by integrating the peak areas of the extracted ions using the Varian MS workstation, version 6.9.2 (Varian Analytical Instruments, Harbour City, CA, USA). The comparative semi-quantifiable results were recorded as the average of triplicate analysis with a mean percentage residual standard deviation (RDS \%) of $8.52 \%$. An autotune was performed at the start of the analysis to ensure that the mass spectrometer was performing optimally.

### 2.4.6 Statistical analysis of food pairing data

### 2.4.6.1 Orthonasal sensory evaluation data

To analyse the sensory data from the focus group the Friedman rank sum test was performed, using a significance level ( $\mathrm{p} \leq 0.05$ ) to determine whether samples were significantly preferred over one another. Follow-up pair wise comparisons were conducted using a Wilcoxon signed rank test and controlling for the Type I errors across
the comparisons at the $\mathrm{p} \leq 0.05$ level using the Fishers Least Squared Difference (LSD) procedure to ascertain where significant differences were. The analysis was performed using SPSS program for Windows (version 19.0, SPSS Inc., Chicago, Il, USA).

### 2.4.6.2 Consumer sensory evaluation of food pairings data

To analyse the consumer hedonic test, the Friedman rank sum was performed, using significance level ( $\mathrm{p} \leq 0.05$ ) to determine whether samples were significantly preferred over one another. Follow-up pair wise comparisons were conducted using a Wilcoxon signed rank test and controlling for the Type I errors across the comparisons at the $\mathrm{p} \leq 0.05$ level using the Fishers Least Squared Difference (LSD) procedure to ascertain where significant differences were. The data from the preference test was analysed using the Pearson's chi squared test and cross-tabulation test. The analysis was performed using SPSS program for Windows (version 19.0, SPSS Inc., Chicago, Il, USA).

### 2.4.6.3 Descriptive sensory and volatile profiling data

Descriptive sensory analysis (Free Choice Profiling (FCP)) data was analysed using Generalised Procrustes Analysis (GPA) using XLSTAT system software, version 2013.4.03, (Addinsoft, Paris, France). GPA has been successfully used to rationalise individual sensory descriptive information from FCP by numerous authors (Rodrigues \& Teixeira, 2013; Lassoued, et al., 2008; Tarea, et al., 2007; Delarue \& Siefffermann, 2004). This method, extracts a common structure from multiple data sets composed of non-identical variables among the data sets (Dijksterhuis, 1996). A consensus plot is
generated that matches the configuration among the multiple data sets by centring, rotating, and adjusting scales of each data set.

APLSR has been used by numerous authors to understand relationships between instrumental (x) and sensory (y) data (Shi et al., 2013; Shiqing et al., 2012; Mitchell et al., 2011). APLSR not only tries to provide solutions for both X and Y variables but also simultaneously attempts to find the 'best' solution of X that will explain the variation of the Y-variable set (Chung et al., 2003). Hence, it compromises between giving weight to the analytical variables and maximising the variance explained (MacFie \& Hedderley, 1993).

The first two factors of the GPA analysis completed $59.88 \%$ of the variance. ANOVA-partial least squares regression (APLSR) was used to interpret the raw data accumulated from the 28 test subjects for the descriptive sensory evaluation and data acquired by volatile analysis. The X-matrix was designed as $0 / 1$ design variables for the individual food samples and the food pairing samples. The Y-matrix was designed as descriptive sensory (orthonasal and retronasal aroma) and volatile analysis variables. The optimal number of components in the APLSR models presented was determined to be four principal components. The validated explained variance for the model constructed was $17 \%$ and the calibrated variance was $45 \%$. APLSR was performed using Unscrambler Software, version 9.7 (CAMO ASA, Trondheim, Norway).

### 2.5 Development of novel ice creams

The development of the novel ice creams is summarised in Figure 2.5.


Figure 2.6 Schematic overview of procedures used in for the development of the ice creams.

### 2.5.1 Ice cream raw materials

Basmati rice, bacon and banana were sourced as described in section 2.4.1. Full fat milk (3 \% fat) (Dunnes stores, Dublin, Ireland), single cream (38 \% fat) (Dunnes stores, Dublin, Ireland), eggs (Dunnes stores, Dublin, Ireland) and caster sugar (Gem Pack Foods Ltd., Dublin, Ireland) were purchased from the local supermarket (Dunnes Stores, Dublin, Ireland). The development of the novel ice creams is summarised in Figure 2.5.

### 2.5.2 Preparation of ice creams

The preparation of the basic ice cream to which the banana, bacon and basmati rice flavours were incorporated was adapted from a preparation set out by McGee (2004) for a premium standard ice cream (Table 2.2). The milk to cream ratio (39.2:35 \% ( $\mathrm{v} / \mathrm{v})$ ) was established to produce a final lipid content of approximately 19 \% (v/v) based on the results of optimised emulsion stability (see section 3.5). Incorporation of the flavours to the ice cream was achieved through preliminary research, where different methods were tested to establish the most efficient method (Appendix D).

Table 2.2. List of ingredients with quantities and percentage of total weight for 1 kg basic ice cream formulation.

| Ingredient | Quantity | Percentage of total weight |
| :---: | :---: | :---: |
| Milk | 392 ml | $39.2 \%(\mathrm{v} / \mathrm{w})$ |
| Cream | 350 ml | $35 \%(\mathrm{v} / \mathrm{w})$ |
| Egg yolk | 108 g | $10.8 \%(\mathrm{w} / \mathrm{w})$ |
| Sugar | 150 g | $15 \%(\mathrm{w} / \mathrm{w})$ |

### 2.5.2.1 Banana and basmati rice ice cream preparation

Raw basmati rice was cooked for 15 minutes in the mixture milk and cream. The rice was then strained, and the cooking liquid reserved. To account for absorption of moisture by the rice and evaporation, the cooking liquid was restored to 1 L with milk. The rice flavoured cooking liquid was added to a mixture of egg yolk and sugar and mixed with a whisk by hand to create a smooth mixture. The mixture was heated to 76 ${ }^{\circ} \mathrm{C}$ for 10 minutes in a pot with constant stirring to create a custard. The custard was placed in a blast chiller (Zanussi Blast Chiller BCF 28kg, Zanussi Professionals, Pordenone, Italy) and chilled to $4{ }^{\circ} \mathrm{C}$. The banana was incorporated into the custard using a hand held blender (Bamix Mono, ESGE Ltd., Mettlen, Switzerland) at full speed for approximately 1 minute. The mixture was placed in an ice cream machine (Musso 4080 Lussino, Musso Lussino, Conegliano, Italy) for approximately 20 minutes until frozen $\left(-4{ }^{\circ} \mathrm{C}\right)$. The ice cream stored in a commercial freezer at $-18^{\circ} \mathrm{C}$ until further use.

### 2.5.2.2 Banana and bacon ice cream preparation

Bacon was cooked in an oven (Rational Combi Dämpfer, Rational UK, Luton, UK) at $190{ }^{\circ} \mathrm{C}$ for 9 minutes and then turned and baked for another 9 minutes. The cooked bacon was cut into small pieces (approximately $2 \mathrm{~cm} \times 3 \mathrm{~cm}$ ) and soaked in the milk and cream mixture for 24 hours. The soaking liquid was strained and the bacon was discarded. The ice cream was made using the bacon flavoured soaking liquid as described in section 2.5.2.1.

### 2.5.3 Determination of preferred flavour concentration ratios

The flavour concentration ranges for banana and basmati rice ice cream ( $B+R$ ) and banana and bacon ice cream $(\mathrm{B}+\mathrm{BN})$ (Table 2.3) were selected based on preliminary screening sensory analysis involving a sensory panel ( $n=6$ ). The intensity of flavour of each ingredient was evaluated, and a range which was considered acceptable (where one ingredient's flavour did not completely overpower or dominate the perceived flavour profile) was selected. Following this, two separate sensory panels ( $\mathrm{n}=34$ ) were conducted to evaluate the taste of the two different sets of ice cream formulations ((B+R 1-5) and (B+BN 1-5)). The sensory panels were made up of staff and students from the School of Culinary Arts and Food Technology and from the School of Food Science and Environmental Health, Dublin Institute of Technology, Dublin.

Table 2.3 Flavour concentration ratios for banana and basmati rice and banana and bacon ice creams.

| Banana and basmati rice ice cream | Banana and bacon ice cream |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Sample | Banana <br> $(\mathbf{g} / \mathbf{k g})$ | Basmati rice <br> $(\mathbf{g} / \mathbf{k g})$ | Sample | Banana <br> $(\mathbf{g} / \mathbf{k g})$ | Bacon <br> $(\mathbf{g} / \mathbf{k g})$ |
| B+R1 | 225 | 150 | B+BN1 | 225 | 150 |
| B+R2 | 225 | 225 | B+BN2 | 225 | 225 |
| B+R3 | 225 | 300 | B+BN3 | 225 | 300 |
| B+R4 | 150 | 300 | B+BN4 | 150 | 300 |
| B+R5 | 150 | 225 | B+BN5 | 150 | 225 |

$\mathrm{B}+\mathrm{R}(1-5)$ : banana and basmati rice ice cream formulations.
$\mathrm{B}+\mathrm{BN}(1-5)$ : banana and bacon ice cream formulations.

Evaluation was carried out in a sensory laboratory under guidelines and conditions according to ISO 8589:2010 (ISO, 2010). Each panellist worked in a single booth under defined conditions of $22{ }^{\circ} \mathrm{C}$ and white light. Approximately 30 g of each ice cream was placed in sample containers coded with three-digit random numbers and were presented in a monadic sequential and randomised order to the panellists and were served at between $-10^{\circ} \mathrm{C}$ and $-13{ }^{\circ} \mathrm{C}$ (Aime et al., 2001). Panellists were instructed to evaluate the taste of the banana and rice ice cream on a 9 point hedonic scale, where $9=$ "like extremely", $5=$ "neither like nor dislike" and $1=$ "dislike extremely". Panellists were instructed to consume mineral water to rinse their mouths between evaluating each sample (Bárcenas et al., 2001). In addition, qualitative data was also recorded; panellists were encouraged to write comments regarding their opinion on the intensity of the banana, rice and bacon flavours in the ice creams. These comments were coded as positive, negative or neutral comments. Evaluation of each ice cream (banana and basmati rice and banana and bacon) took place on separate days.

### 2.5.4 Salt (sodium chloride) content of preferred ice creams

### 2.5.4.1 Moisture content measurement

Moisture content of banana and basmati rice (B+RI), banana and bacon (B+BNI, banana (Control 1) and plain (Control 2) ice creams was determined by the AOAC method 925.098 (AOAC, 1990). Ten grams samples of each ice cream were weighed and then dried in an oven (Universal Oven, Memmert, Schwabach, Germany) at $105{ }^{\circ} \mathrm{C}$ for 24 hours. Following this, the samples were removed from the oven, cooled in a desiccator to room temperature and weighed. Percentage moisture (Equation 2.6) was calculated using the following equation as described by Bradley (2010);
$\%$ Moisture $=\frac{\text { weight } \operatorname{loss}(g)}{\text { wet weight }(g)} \times 100$
Equation 2.6
2.5.4.2 Preparation of samples for atomic absorption spectrometry

Analytical grade reagents were used throughout. Distilled, deionized water (DDW) from a Milli-Q water purification system (Millipore, Bedford, MA, USA), was used for the preparation of the solutions. All containers and glassware were washed with $69 \%$ nitric acid $\left(\mathrm{HNO}_{3}\right)$ and rinsed with DDW before use. Fifty mL of $\mathrm{HNO}_{3}$ (69 \%) was added to 3 g of dried ice cream in a glass beaker and the contents was swirled gently. The solution was then heated to $120^{\circ} \mathrm{C}$ on a hotplate. After approximately 40 minutes, the content of beaker was reduced to near dryness producing a clear orange solution. The beaker was removed from the hotplate and allowed to cool to room temperature. During cooling, 50 mL of $\mathrm{HNO}_{3}$ was added to obtain a final digestate solution of approximately 64 mL . Fifteen mL of the digestate solution was pipetted into a 100 mL volumetric flask and topped up with DDW.

### 2.5.4.3 Atomic absorption spectrometry

All measurements were carried out using a model AAS 6 Varion atomic absorption spectrometer (Analytik Jena AG, Jena, Germany). A sodium hollow cathode lamp (Photron pty Ltd., Melbourne, Australia) operating at 7 mA was used as the radiation source. The atomisation was carried out in an air-acetylene flame. The acetylene flow rates and the burner height were adjusted in order to obtain the maximum absorbance signal. High purity argon was used as a carrier gas. Three replicates of each sample were prepared and the experiment was repeated three times. A
calibration curve using NaCl as a standard was constructed and used for quantification of NaCl content. The standard curve had an $R^{2}$ value of $0.999 . \mathrm{NaCl}$ concentration was calculated using the following:
salt conc. $=\left(\frac{A}{e l}\right) \times 2.5$
Equation 2.7

Where A is the absorbance, e is the slope, 1 is the path length and results were expressed as $\mathrm{g} / 100 \mathrm{~g}$.

### 2.5.5 pH measurement

 (Control 1) and plain (Control 2) ice creams at room temperature were determined using an Orion Model 520A pH metre (AGB Scientific Ltd., Dublin, Ireland). The pH of 30 mL of ice cream sample was measured at $20^{\circ} \mathrm{C}$. Three replicates of each sample were prepared and the experiment was repeated three times.

### 2.5.6 Evaluation of consumer acceptance and market suitability

To evaluate the acceptability of the ice cream products to two selected markets, two groups of subjects were selected representing each separate markets; a general retail market (panel A) and a culinary educated market (panel B). The rationale for assessing the two ice cream samples with two sensory panels representing different markets was based on findings from the focus groups (section 5.2), where certain participants with no professional culinary background displayed partial neophobic behaviour towards some of the food pairings. In contrast, participants with professional culinary education
appeared to display neophilic behaviour towards some of the food pairings. In addition to this, the naturally lower levels of salt in banana and basmati rice ice cream than in banana and bacon ice cream also allowed for assessment of two contrasting products in light of this partial neophobic behaviour.

Panel A was made up of staff and students from the School of Culinary Arts and Food Technology and from the School of Food Science and Environmental Health, Dublin Institute of Technology, Dublin. Panel B was made up solely of culinary students from the School of Culinary Arts and Food Technology, Dublin Institute of Technology, Dublin. In total, 171 females and 133 males who ranged in age from 18 to 45 years of age evaluated the taste acceptability of the two ice cream samples ( $B+R$ and $B+B N)$ in four separate sensory panels, with 76 participants in each $(n=304)$. The ice creams $(B+R$ and $B+B N)$ were evaluated separately on different days, and also separately by each panel (panel A and panel B) on different days (4 separate days in total). Sensory evaluation was conducted as described in section 2.5.3. Panellists were instructed to evaluate the taste of the banana and rice ice cream on a 9 point hedonic line scale, where $9=$ "like extremely", $5=$ "neither like nor dislike" and $1=$ "dislike extremely".

### 2.5.7 Statistical analysis of novel ice cream data

### 2.5.7.1 Determination of optimum flavour concentrations data

The Friedman rank sum was performed, using a significance level ( $p \leq 0.05$ ) to determine whether samples were significantly preferred over one another. Follow-up pair wise comparisons were conducted using a Wilcoxon signed rank test and
controlling for the Type I errors across these comparisons at the 0.05 level were controlled using the Fishers Least Squared Difference (LSD) procedure to ascertain where significant differences where. The analysis was performed using SPSS program for Windows (version 19.0, SPSS Inc., Chicago, Il, USA).

### 2.5.7.2 Salt and pH measurement data

To analyse the salt and pH measurement data from the four separate ice cream samples, one-way analysis of variance (ANOVA) was performed, using a significance level of $\mathrm{p} \leq 0.05$. This was followed by Fisher's least significant differences (LSD) test to ascertain where significant differences were. The analysis was performed using SPSS program for Windows (version 19.0, SPSS Inc., Chicago, Il, USA).
2.5.7.3 Consumer acceptance and market suitability evaluation data

The consumer evaluation data from the four separate sensory panels, was analysed as in section 2.5.7.2.

## Chapter 3

### 3.1 General introduction

This chapter involved the optimisation of the formation and stability of an oil in water emulsion food model system containing lecithin, xanthan gum and sunflower oil. Even in a simple model food system a vast array of factors can influence formation and storage stability (Ribeiro et al., 2011). Obtaining knowledge of the formation and stability properties of emulsion ingredients is vital for the formulation of emulsion based model systems. This also enables the further investigation of other culinary mechanisms of phenomena like flavour release from such model systems. Response surface methodology and nonlinear regression were applied as statistical tools to (1) model and optimise the emulsion ingredient levels (lecithin $\left(x_{1}\right)$, xanthan gum $\left(x_{2}\right)$ and sunflower oil $\left.\left(x_{3}\right)\right)$ leading to maximum emulsion storage stability ratio and minimum mean droplet diameter and (2) study the linear, quadratic and interactive effects of emulsion ingredient concentration on storage stability ratio and mean droplet diameter.

### 3.2 Fitting of the response surface models and repeatability

The independent variables and experimental design factor setting for the Box Behnken design including the mean values and corresponding standard deviations of the two responses: emulsion storage stability ratio (SR) and mean droplet diameter (DS) for each experimental condition are illustrated in Table 3.1. The regression coefficients and analysis of variance of the coded independent variables for SR are presented in Table 3.2. Besides showing the optimal conditions for maximum SR and minimum DS, the mathematical model was able to identify and describe significant effects of the independent variables, their relative sensitivity and some interesting interactions between the variables on both responses.

Table 3.1 Experimental design with level of factors (low ( $-\alpha$ ), medium ( $\alpha$ ) and high $(+\alpha)$ ), mean values and standard deviation for responses according to the Box Behnken design.

| Run | Sunflower oil (\% v/v) | Lecithin (\% w/v) | $\begin{gathered} \text { Xanthan } \\ \text { gum } \\ (\% \mathrm{w} / \mathrm{v}) \end{gathered}$ | $\begin{aligned} & \hline \text { SR } \\ & (\%) \end{aligned}$ |  | $\begin{gathered} \hline \mathrm{DS} \\ (\mu \mathrm{~m}) \end{gathered}$ |  | SD |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 15 | 5 | 0.01 | 67.8 | $\pm 6.0$ | 5.74 | $\pm$ | 0.24 |
| 2 | 10 | 3 | 0.3 | 100 | $\pm 0$ | 4.94 | $\pm$ | 0.11 |
| 3 | 10 | 3 | 0.01 | 88.9 | $\pm 1.0$ | 5.4 | $\pm$ | 0.05 |
| 4 | 15 | 3 | 0.16 | 51.0 | $\pm 2.8$ | 5.31 | $\pm$ | 0.05 |
| 5 | 10 | 1 | 0.16 | 92.1 | $\pm 1.6$ | 5.24 | $\pm$ | 0.04 |
| 6 | 15 | 1 | 0.01 | 81.3 | $\pm 1.8$ | 5.13 | $\pm$ | 0.08 |
| 7 | 10 | 5 | 0.16 | 54.3 | $\pm 2.8$ | 5.08 | $\pm$ | 0.18 |
| 8 | 20 | 3 | 0.01 | 71.0 | $\pm 1.1$ | 6.05 | $\pm$ | 0.35 |
| 9 | 20 | 5 | 0.16 | 62.9 | $\pm 2.0$ | 5.26 | $\pm$ | 0.29 |
| 10 | 15 | 5 | 0.3 | 100 | $\pm 0$ | 4.69 | $\pm$ | 0.21 |
| $11^{\text {c }}$ | 15 | 3 | 0.16 | 48.5 | $\pm 2.9$ | 5.48 | $\pm$ | 0.15 |
| $12^{\text {c }}$ | 15 | 3 | 0.16 | 50.7 | $\pm 2.0$ | 5.41 | $\pm$ | 0.09 |
| 13 | 20 | 1 | 0.16 | 100 | $\pm 0$ | 5.28 | $\pm$ | 0.12 |
| 14 | 20 | 3 | 0.3 | 100 | $\pm 0$ | 4.91 | $\pm$ | 0.12 |
| 15 | 15 | 1 | 0.3 | 100 | $\pm 0$ | 4.96 | $\pm$ | 0.12 |

${ }^{\mathrm{c}}$ centre points.
SD: standard deviation.
SR: emulsion storage stability ratio.
DS: mean droplet diameter.

The $R^{2}$ statistic indicated that the response surface model accounted for $89 \%$ of the variation for SR and $86 \%$ for DS. When the residual plots were investigated, they appeared to follow a normal distribution and to be independent from any of the variables. The repeatability of the experimental procedure was evaluated by calculating the relative standard deviation percentage (RSD \%) of three replicates of each experimental condition. The RSD \% for SR ranged from 0 to 8.8, with a RSD \% average of 2.6, while the RSD \% for average DS ranged from 0.2 up to 5.7 , with a RSD
\% average of 2.6 , indicating that the replication within each experimental condition was good.

### 3.3 Stability of emulsions

Polynomial parameter estimates for xanthan gum indicate that linear ( $x_{3}$ ), quadratic $\left(x_{3}{ }^{2}\right)$ and interactive ( $x_{1} \cdot x_{3}$ and $x_{2} \cdot x_{3}$ ) effects were significantly influential ( $\mathrm{p} \leq$ 0.01 ) on SR (Table 3.2). These effects proved to positively influence SR. Additionally, the magnitude of xanthan gum's influence on SR can be seen in the Pareto effects chart (Figure 3.1).

Table 3.2 Regression coefficients and analysis of variance of coded independent variables for stability ratio (SR).

| Regression coefficients | SR |  | SE |
| :---: | :--- | :--- | :--- |
| $\beta_{0}$ | $50.05^{* * *}$ | $\pm$ | 37.89 |
| $\beta_{1}$ | -0.85 | $\pm$ | -1.02 |
| $\beta_{2}$ | $-10.40^{* * *}$ | $\pm$ | -12.51 |
| $\beta_{3}$ | $11.38^{* * *}$ | $\pm$ | 14.07 |
| $\beta_{12}$ | -1.13 | $\pm$ | -0.93 |
| $\beta_{13}$ | $4.48^{* * *}$ | $\pm$ | 3.92 |
| $\beta_{23}$ | $3.38^{* *}$ | $\pm$ | 2.96 |
| $\beta_{1}{ }^{2}$ | $15.70^{* * *}$ | $\pm$ | 13.00 |
| $\beta_{2}{ }^{2}$ | $12.97^{* * *}$ | $\pm$ | 10.76 |
| $\beta_{3}{ }^{2}$ | $24.26^{* * *}$ | $\pm$ | 20.12 |
| $\boldsymbol{R}^{2}$ | 0.89 |  |  |
| Regression (P-value) | $<2.2 \mathrm{e}-16^{* * *}$ |  |  |
| Lack of fit (P-value) | $<2.2 \mathrm{e}-16^{* * *}$ |  |  |

$\beta_{1}$ : estimated regression for the linear effects.
$\beta_{l}{ }^{\varphi}$ : estimated regression coefficients for the quadratic effects.
$\beta_{u}$ : estimated regression coefficients for the interaction effects.

* significant ( $\mathrm{p} \leq 0.05$ ).
${ }^{*}$ ** highly significant ( $\mathrm{p} \leq 0.01$ ).
${ }^{* * *}$ very highly significant ( $\mathrm{p} \leq 0.001$ )
SE: standard error.
SR: emulsion storage stability ratio.


Figure 3.1 Pareto effects chart for stability ratio (SR).
Line represents cumulative percentage total.

The linear ( $x_{3}$ ) and quadratic (squared polynomial) terms ( $x_{3}{ }^{2}$ ) were located in the upper end of the Pareto effects chart implying they were highly influential on SR, while the interactive terms were in the lower end. The highly influential linear effect for xanthan gum concentration implies that there was linearity in the relationship between xanthan gum concentration and SR. While, the highly influential quadratic effect indicates that there was a curvilinear relationship between xanthan gum concentration and SR, this can be seen in Figures $3.2 \& 3.3$. These results suggest that varying the concentration of xanthan gum was the most influential variable on SR, and additions of xanthan gum resulted in an increase in SR, consequently enhancing its resistance to gravitational separation. Nevertheless, when xanthan gum concentrations were initially low ( $0.01 \% \mathrm{w} / \mathrm{v}$ ), the SR range was $70-80 \%$ (Figures $3.2 \& 3.3$ ). However, at intermediate xanthan gum concentrations ( $0.16 \% \mathrm{w} / \mathrm{v}$ ), SR further decreased to 50-60
\%. Once levels of xanthan gum passed this intermediate point (> 0.16 \% w/ v), SR subsequently increased to $100 \%$.


Figure 3.2 Response surface plots for emulsion stability as a function of xanthan gum and lecithin concentration when sunflower oil is fixed at 0.08 .


Figure 3.3 Response surface plots for emulsion stability as a function of xanthan gum and sunflower oil concentration when lecithin is fixed at 0.44.

Dickinson (2009) found that at low concentrations, added hydrocolloids (such as xanthan gum) can have a destabilising influence on emulsion stability due to a mechanism known as depletion flocculation. This mechanism is induced by the excess non-absorbing hydrocolloid and/or surfactant forming micelles. In fact, xanthan gum is known as a common depletion flocculant, with specific critical concentrations for such mechanisms being reported (Sun et al., 2007). However, Hemar et al. (2001) observed that although increases in xanthan gum content caused more extensive flocculation of droplets, overall emulsion stability was subsequently improved. Similarly, Ye et al. (2004) obtained accelerated droplet flocculation with emulsion re-stabilisation occurring once xanthan gum concentration exceeded a critical concentration ( $0.12 \% \mathrm{w} / \mathrm{v}$ ). This stabilising effect of xanthan gum has been previously attributed to its ability to increase the viscosity of the continuous phase, thereby minimising droplet mobility and decreasing droplet collision numbers (Tsaliki et al., 2004; Dickinson, 2003). Hence, it may be suggested that a critical concentration of xanthan gum ( $0.16 \% \mathrm{w} / \mathrm{v}$ ) was present, at which a destabilising effect from xanthan gum was prevalent. Once surpassed, the apparent viscosity of the continuous phase was increased, thereby reducing the mobility of the emulsion droplets inhibiting aggregation or coalescence (Sworn, 2000).

Lecithin was also found to have significant $(\mathrm{p} \leq 0.05)$ linear $\left(x_{2}\right)$, quadratic $\left(x_{2}{ }^{2}\right)$ and interactive $\left(x_{2} \cdot x_{3}\right)$ effects on SR (Table 3.2). Although lecithin was not found to be as influential as xanthan gum on SR, the magnitude and importance of its influence on SR is clear from the Pareto effects chart (Figure 3.1). The linear $\left(x_{2}\right)$ and quadratic $\left(x_{2}{ }^{2}\right)$ effects of lecithin are positioned high on the Pareto effects chart with a relatively high magnitude of standardised effect. These linear and quadratic effects demonstrate that there was a curvilinear effect present. Thus, varying lecithin concentration had an
important bearing on SR. Figures 3.2 \& 3.4 suggest that this influence was negative, higher levels of lecithin (> $2.5 \% \mathrm{w} / \mathrm{v}$ ) appeared to have a destabilising effect. Therefore, a similar depletion flocculation mechanism (as was the case with xanthan gum), where excess non-absorbed surfactant promoted phase separation may have been a reason for this phenomena.

The interactive effect of sunflower oil with xanthan gum ( $\mathrm{x}_{1} \cdot \mathrm{x}_{3}$ ) was found to be significant ( $\mathrm{p} \leq 0.05$ ) with respect to SR (Table 3.2). The quadratic effect of sunflower oil ( $x_{1}{ }^{2}$ ) was also found to be significant ( $\mathrm{p} \leq 0.05$ ), which indicates that there was a certain degree of curvilinear effects present regarding sunflower oil's relationship with SR (Table 3.2). Changes in sunflower oil concentration $\left(x_{l}\right)$ had less of an influence in comparison with the other design variables on SR (Figures 3.2 \& 3.4).


Figure 3.4 Response surface plot for emulsion stability as a function of lecithin and sunflower oil concentration when xanthan is fixed at -0.27 .

It is worth noting that intermediate levels of all independent variables produced the least desirable SR in the present study. Overall, the graphical results seem to indicate that the optimal SR conditions are when high xanthan gum, low lecithin and high sunflower oil are used. SR kinetic plots (triplicate samples) through storage time for experimental conditions 4 and 8 are depicted in Figures 3.5 and Figure 3.6 respectively. In the case of experimental condition 4 (Figure 3.5), initially, SR remained at $100 \%$ for day 0 and day 1 , however, a sharp decline can be seen from day 1 to day 7 $(59.36 \%)$ with a levelling out or "plateau" period to day 14 indicating little or no change. A similar behaviour can be seen for experimental condition 8 (Figure 3.6) with SR declining sharply from $100 \%$ on day 0 to $71 \%$ on day 1 and levelling out throughout the remainder of storage time.


Figure 3.5 Stability ratio kinetics plot for experimental condition 4.


Figure 3.6 Stability ratio kinetics plot for experimental condition 8 .

### 3.4 Emulsion mean droplet diameter

Statistical significant parameters ( $\mathrm{p} \leq 0.05$ ) for the secondary model building of the DS response are listed in Table 3.3. The results for initial DS $\left(R_{0}\right)$ showed that all linear, quadratic and interactive variables for sunflower oil $\left(x_{1}\right)$, lecithin ( $x_{2}$ ) and xanthan gum $\left(x_{3}\right)$ significantly influenced $(\mathrm{p} \leq 0.05) R_{0}$ (except for the quadratic effect of xanthan gum $\left(x_{3}{ }^{2}\right)$ and the interactive effect $\left(x_{1} x_{3}\right)$ ). The lack of a significant quadratic effect for xanthan gum and the presence of a significant linear effect ( $R_{0 x 3}$ ) are depicted in Figure 3.7, where a clear linearity can been seen between the relationship of xanthan gum concentration and initial DS $\left(R_{0}\right)$. Moreover, this linear effect ( $R_{0 x 3}$ ) and interactive effect with lecithin ( $R_{0 x 2} \times x_{3}$ ) were found to be negative.

Table 3.3 Regression coefficients and analysis of variance of coded independent variables for mean droplet diameter (DS).

| Regression Coefficients | DS | SE |
| :---: | :---: | :---: |
| Asym $_{0}$ | 5.41 | $\pm 0.04$ |
| Asym $_{x 3}$ | -0.64 | $\pm 0.04$ |
| Asym $_{x 3 \times 2}$ | -0.11 | $\pm 0.05$ |
| $R_{00}$ | 4.59 | $\pm 0.04$ |
| $R_{0 x 1}$ | 0.12 | $\pm 0.03$ |
| $R_{0 x 2}$ | -0.21 | $\pm 0.03$ |
| $R_{0 x 3}$ | -0.30 | $\pm 0.03$ |
| $R_{0 x 1^{2}}$ | 0.30 | $\pm 0.04$ |
| $R_{0 x 2} 2^{2}$ | -0.11 | $\pm 0.04$ |
| $R_{0 x 1 \times x 2}$ | 0.07 | $\pm 0.03$ |
| $R_{0 x 2 \times 3}$ | -0.27 | $\pm 0.04$ |
| $l r c_{0}$ | -1.4 | $\pm 0.2$ |
| $l r c_{x 3}$ | 1.51 | $\pm 0.2$ |
| $l r c_{x 2} 2^{2}$ | -1.2 | $\pm 0.2$ |
| $l r c_{x 3}{ }^{2}$ | 1.2 | $\pm 0.2$ |
| $l r c_{x 3 \times x 2}$ | -0.38 | $\pm 0.16$ |
| $l r c_{x 3 \times x 1}$ | -1.8 | $\pm 0.3$ |
| $R^{2}$ | 0.86 |  |

All parameter estimates significant ( $\mathrm{p} \leq 0.05$ ).
SE: standard error.
Subscripts in parameter estimate name indicate the polynomial dependence term in the final secondary model building.


Figure 3.7 Response surface plots for mean droplet diameter (DS) estimated parameter $R_{0}$ (Initial DS) as a function of xanthan gum and sunflower oil concentration when lecithin was fixed at -0.37 .

All effects of lecithin were found to negatively influence $R_{0}$ (Table 3.3). The presence of a significant $(\mathrm{p} \leq 0.05)$ quadratic effect $\left(\mathrm{R}_{0 \times 2}{ }^{2}\right)$ implies that there was curvature in the relationship between lecithin concentration and initial DS $\left(R_{0}\right)$ which can be seen in Figures $3.8 \mathrm{a} \& \mathrm{~b}$. High concentrations of lecithin and xanthan gum produced the most desirable $R_{0}$. It can be seen that as lecithin and xanthan gum concentrations increased, $R_{0}$ subsequently decreased. The influential properties of lecithin in the reduction of initial DS $\left(R_{0}\right)$ in this study would be expected. A primary role of an emulsifier is to migrate to the interface of the newly formed droplets, form a protective layer which prevents aggregation, and reduce the interfacial tension, therefore stabilising against coalescence (McClements, 2005). However, in a study by Tirok et al. (2001), lecithin concentration was found to not be an important influential factor on the emulsion stability and mean droplet diameter. Sunflower oil was found to have a
positive effect on $R_{0}$, thus, addition of sunflower oil caused an increase in $R_{0}$. Figure 3.8a shows that intermediate levels of sunflower oil ( $15 \% \mathrm{v} / \mathrm{v}$ ) resulted in a more favourable initial DS $\left(\mathrm{R}_{0}\right)$.
(a)

(b)


Figure 3.8 Response surface plots for mean droplet diameter (DS) estimated parameter $R_{0}$ (Initial DS) as a function of (a) lecithin and sunflower oil concentration when xanthan gum was fixed at 0.44 and (b) xanthan gum and lecithin concentration when sunflower oil was fixed at 0.03 .

It was found that xanthan gum proved to be the critical influential variable on the speed of DS growth ( $l r c$ ). The linear $\left(l r c_{x 3}\right)$, quadratic $\left(l r c_{x 3}{ }^{2}\right)$ and interactive ( $\operatorname{lrc_{x3x}}$ ) and $\left(l r c_{x 3 \cdot x 2}\right)$ effects of xanthan gum were significantly influential $(\mathrm{p} \leq 0.05)$ on lrc. It is clear from Figure 3.9 that higher levels of xanthan gum ( $0.16-0.3 \% \mathrm{w} / \mathrm{v}$ ) in combination with low levels of sunflower oil ( $10-15 \% \mathrm{v} / \mathrm{v}$ ) appear to have had the effect of increasing the rate of DS growth.


Figure 3.9 Response surface plots for mean droplet diameter (DS) estimated parameter lrc (log of the first order apparent rate constant) as a function of xanthan gum and sunflower oil concentration when lecithin was fixed at 0.32 .

The quadratic effect of lecithin $\left(l r c_{x 2}{ }^{2}\right)$ also had a significant influence ( $\mathrm{p} \leq 0.05$ ) on the lrc which is clear from the curvature present in Figure 3.10. Xanthan gum in combination with lecithin ( $\operatorname{lrc}_{x 3 \times 2}$ ) was found to negatively influence lrc, thus, producing a desirable reduction in DS growth rate. When both xanthan gum and lecithin were at lower levels ( $\leq 0.16 \% \mathrm{w} / \mathrm{v}$ and $\leq 1.25 \% \mathrm{w} / \mathrm{v}$ respectively), a desirable lrc
resulted (Figure 3.10). However, intermediate levels of lecithin (2.5 \% w/v) in combination with high levels of xanthan gum ( $0.24-0.3 \% \mathrm{w} / \mathrm{v}$ ) proved to produce an undesirable increase in lrc.


Figure 3.10 Response surface plots for mean droplet diameter (DS) estimated parameter lrc (log of the first order apparent rate constant) as a function of xanthan gum and lecithin concentration when sunflower oil was fixed at 0.81 .

Figure 3.11 appears to indicate that intermediate levels of lecithin ( $2.5 \% \mathrm{w} / \mathrm{v}$ ) regardless of sunflower oil concentrations resulted in an increase in the rate of DS growth. Hence, the highest rate DS growth was at an intermediate lecithin concentration ( $2.5 \% \mathrm{w} / \mathrm{v}$ ).


Figure 3.11 Response surface plots for mean droplet diameter (DS) estimated parameter lrc (log of the first order apparent rate constant) as a function of lecithin and sunflower oil concentration when xanthan gum was fixed at 0.56 .

In terms of final mean droplet diameter (Asym), xanthan gum (Asym ${ }_{x 3}$ ) proved to have a significant influence ( $\mathrm{p} \leq 0.05$ ) on Asym (Table 3.3), increasing xanthan gum concentration reduced the final DS in the emulsions. Additionally, the combination of xanthan gum and lecithin $\left(\operatorname{Asym}_{x 3 \times 2}\right)$ also had a significant influence $(\mathrm{p} \leq 0.05)$ in reducing the final DS. A lack of significant quadratic variables indicated that the response surface was linear, with no maxima or minima found in this study.

The droplet diameter distribution over storage time of the two different experimental conditions (condition 10 and 11) are shown in Figures 3.12a \& b respectively. Two contrasting behaviours can be seen in these two plots. A relatively narrow distribution curve with a small droplet diameter range and little variation over storage time is depicted in Figure 3.12a. On the other hand, it can be seen in Figure
3.12b, that the distribution curve on day 0 is relatively narrow, however as time increases, the distribution curve becomes wider with a slight shift towards the right indicating that the droplet diameter increased and also that diameter size became less homogenous.


Figure 3.12 Droplet diameter distribution density plot for (a) emulsion condition 10 and (b) condition 11 .

```
day 0 - day 1 - day 7 - day 14
```

Figure 3.13a depicts a kinetic plot of DS (triplicate samples) through storage time for experimental condition 7. It would appear that there was a somewhat linear relationship between DS and storage time for experimental condition 7, with DS proportionally increasing through storage time. However, Figure 3.13b depicts contrasting behaviour for condition 12 in comparison to experimental condition 7. A more complicated nonlinear relationship between DS and storage time appears to have been present. After 7 days of storage a "plateau" was reached, in which DS "levels out" with little or no change. This behaviour appears to be present for the majority of the experimental conditions.
(a)

(b)


Figure 3.13 Mean droplet diameter kinetics plot for (a) experimental condition 7 and (b) experimental condition 12 .

Regarding DS as a whole, the results suggest that xanthan gum was the key influential variable in reducing initial $\mathrm{DS}\left(R_{0}\right)$, minimising droplet growth rate (lrc) and reducing the final DS (Asym). Xanthan gum's ability for minimising droplet mobility can provide the primary benefit of delaying droplet-droplet contact (Ye et al., 2004). A secondary benefit of xanthan gum's ability to minimising droplet mobility is that it can provide sufficient time for the emulsifier (lecithin) to migrate to the droplet interface (Cao et al., 1990). Krstonosic et al. (2009) observed that an increase in xanthan gum concentration led to a decrease in mean droplet diameter, however, other studies observed that the effective diameter of the droplets in the emulsion increased because of flocculation caused by xanthan gum (Ye et al., 2004; Hemar et al., 2001).

### 3.5 Multi Criteria Optimisation and validation of optimal conditions

Multi criteria optimisation was employed to find a compromise between the two response variables in the study using the library MCO (R 2.14.1 software package). A Pareto front plot for the two optimised responses was produced after generating a population of 1,000 individual optimisations (Figure 3.13). The lack of any curvature in the Pareto front plot, suggests that the optimisation process resulted in no compromise being achieved between the two responses. Thus, any optimisation that attempts to improve the emulsion properties from a particular start up point will result in a benefit in one of the responses that will cause a directly proportional loss in the other response. Therefore, any decrease in DS resulted in a corresponding linearly correlated decrease in SR and vice versa. Hence, a 50/50 weighting was found to be the best condition and was tested to show the ability of the model to optimise the product. These experimental measures were used to follow the validation study.


Figure 3.14 Pareto front plot showing the simultaneous optimisation of the droplet diameter and the stability ratio.

Stability and average droplet diameter analysis were carried out at the optimal conditions to verify the model. A validation study was conducted employing one set of the optimal conditions from the Pareto front (sunflower oil ( $\mathrm{x}_{1}$ ) $19.02 \%$, lecithin ( $\mathrm{x}_{2}$ ) $1.2 \%$, xanthan gum ( $\mathrm{x}_{3}$ ) $0.28 \%$ ). The predicted values were $\mathrm{SR} \leq 100 \%$ and DS 4.49 $\mu \mathrm{m}$, the experimental values were SR $100 \%$ and DS $4.35 \mu \mathrm{~m}$. It should be noted that both the asymptotic nonlinear regression and polynomial regressions equation were only a statistical empirical model in the selected ranges. An alternative modelling emulsion droplet diameter distribution using the Sauter diameter ( $\mathrm{D}^{32}$ ) was explored, however due to variability amongst samples, a low $R^{2}$ value was achieved and this could not be further examined.

Droplet diameter is a key characteristic of emulsion stability, contributing greatly to the physical stability and organoleptic properties of emulsions, where large globules tend to coalesce faster than the small ones as in Ostwald ripening (McClements, 2005). Stoke's law states that the velocity at which a droplet moves is proportional to the square of the droplet size radius (Dłużewska et al., 2006). Thus, in theory, reducing the size of the droplets should enhance the stability of an emulsion to gravitational separation (Huang et al., 2001). To achieve a decrease in the coalescence rate by a factor of 10-100, a decrease in mean globule diameter by a factor of two is required (Bergenstahl \& Claesson, 1997). It was found that a decrease in droplet diameter by $0.02 \mu \mathrm{~m}$ resulted in a decrease in emulsion stability by $8 \%$, which translates to a coalescence rate with a factor of 400 (Figure 3.14).

### 3.6 Conclusions

Results show that both xanthan gum and lecithin were crucial components for extending the emulsions shelf life to 14 days at ambient room temperature. A critical concentration for xanthan gum ( $0.16 \% \mathrm{w} / \mathrm{v}$ ) was established; at which emulsion storage stability was at its lowest, once exceeded stability was enhanced. Both initial and final droplet diameters were minimised with increasing concentrations of xanthan gum. Although increasing the lecithin concentration promoted desirable mean droplet diameters, it also induced emulsion instability, thus, lower levels (< $2.5 \% \mathrm{w} / \mathrm{v}$ ) were found to be optimal. While, sunflower oil was less influential on emulsion storage stability and mean droplet diameter, its interaction with xanthan gum and lecithin, at intermediate levels of sunflower oil ( $15 \% \mathrm{v} / \mathrm{v}$ ), proved to induce undesirable results for both responses. Optimum conditions were found to be sunflower oil $19.02 \% \mathrm{v} / \mathrm{v}$, soy lecithin $1.2 \% \mathrm{w} / \mathrm{v}$ and xanthan gum $0.28 \% \mathrm{w} / \mathrm{v}$. The optimisation process resulted in no compromise being achieved between the two responses, a decrease in droplet diameter by $0.02 \mu \mathrm{~m}$ resulted in a decrease in emulsion stability by $8 \%$. The knowledge of the stability properties developed in this work is essential for the formulation of emulsion based food products such as sauces, ice cream and salad dressings. It is also important to note the limitations of an emulsion model system especially in terms of the application of findings to more complex food systems such as ice cream. Emulsification is a critical initial stage in ice cream production. Hence, producing the most stable emulsions is an important foundation step in the production of ice cream. However, as outlined in section 1.3.3, there are numerous ingredients (suspension, emulsion, foam etc.) and processing steps involved in transforming the initial food emulsion to a more complex food dispersion (ice cream). The model developed in this work did not completely represent an ice cream product.

## Chapter 4

### 4.1 General introduction

In this chapter, the optimised food emulsion model system was investigated further to examine which physicochemical conditions would result in maximum volatile emissions of isoamyl acetate and furfuryl acetate. Both acetates are commonly used banana flavour compounds, eliciting fruity notes (Burdock, 2010; Yilmaztekin et al., 2008). The production of foods/dishes in a kitchen or in a food manufacturing plant involves manipulation of the many physicochemical characteristics of the product, which can influence the release of volatile molecules during gustation (Traynor et al., 2012). For example, many foods contain significant levels of added sodium chloride $(\mathrm{NaCl})$, both to deliver flavour and act as a preservative (Mitchell et al., 2011). Information on the effect of the characteristics of food matrices with respect to flavour volatile release is necessary. RSM was employed to (1) model and optimise the pH conditions and salt concentration ( NaCl ) leading to maximum volatile emission, and (2) to study the linear, quadratic and interactive effects of the experimental parameters on the volatile emission.

### 4.2 Fitting of the response surface models and repeatability

The independent variables and experimental design factors for the Central Composite Faced Centred experimental design (CCF), including the mean values and corresponding standard deviations of the two responses: isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ and furfuryl acetate $\left(\mathrm{Y}_{2}\right)$ emission are outlined in Table 4.1.

Table 4.1 Experimental design with independent variables ( $\mathrm{x}_{\mathrm{i}}$ ) (low ( $-\alpha$ ), medium ( $\alpha$ ) and high $(+\alpha))$ and response variables $\left(\mathrm{y}_{\mathrm{j}}\right)$ according to Central Composite Face Centred (CCF).

| Run | Independent variables |  |  |  | Response Variables |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Isoamyl acetate (ppm) | Furfuryl acetate (ppm) |  | $\begin{gathered} \text { Salt } \\ (\mathrm{w} / \mathrm{v} \%) \end{gathered}$ | Isoamyl acetate (ppm) ( $\mathbf{Y}_{1}$ ) | $\begin{gathered} \hline \text { RSD } \\ \% \end{gathered}$ | $\begin{gathered} \text { Furfuryl } \\ \text { acetate } \\ (\mathbf{p p m})\left(\mathbf{Y}_{2}\right) \end{gathered}$ | $\begin{gathered} \hline \text { RSD } \\ \% \end{gathered}$ |
| 1 | 50 | 20 | 5 | 0.1 | 150.2 | 7.5 | 136.66 | 10.8 |
| 2 | 90 | 20 | 5 | 0.1 | 284.5 | 5.0 | 246.29 | 6.1 |
| 3 | 50 | 30 | 5 | 0.1 | 106.7 | 3.8 | 242.59 | 6.1 |
| 4 | 90 | 30 | 5 | 0.1 | 96.8 | 18.0 | 81.10 | 30.0 |
| 5 | 50 | 20 | 7 | 0.1 | 73.2 | 7.4 | 316.64 | 14.0 |
| 6 | 90 | 20 | 7 | 0.1 | 245.3 | 7.2 | 174.60 | 9.1 |
| 7 | 50 | 30 | 7 | 0.1 | 20.8 | 2.3 | 40.87 | 9.6 |
| 8 | 90 | 30 | 7 | 0.1 | 93.0 | 0.9 | 77.23 | 1.5 |
| 9 | 50 | 20 | 5 | 2 | 110.9 | 1.4 | 276.91 | 2.2 |
| 10 | 90 | 20 | 5 | 2 | 277.9 | 4.6 | 207.03 | 5.6 |
| 11 | 50 | 30 | 5 | 2 | 140.8 | 4.1 | 262.06 | 5.9 |
| 12 | 90 | 30 | 5 | 2 | 269.8 | 6.0 | 281.69 | 7.4 |
| 13 | 50 | 20 | 7 | 2 | 124.4 | 2.3 | 168.84 | 3.5 |
| 14 | 90 | 20 | 7 | 2 | 364.6 | 3.6 | 133.49 | 4.2 |
| 15 | 50 | 30 | 7 | 2 | 40.4 | 13.8 | 78.38 | 35.9 |
| 16 | 90 | 30 | 7 | 2 | 309.3 | 6.5 | 253.63 | 7.9 |
| 17 | 50 | 25 | 6 | 1.05 | 24.1 | 7.9 | 48.08 | 29.0 |
| 18 | 90 | 25 | 6 | 1.05 | 287.8 | 2.3 | 241.22 | 2.8 |
| 19 | 70 | 20 | 6 | 1.05 | 194.0 | 21.5 | 101.41 | 28.7 |
| 20 | 70 | 30 | 6 | 1.05 | 1980.0 | 7.2 | 317.92 | 9.5 |
| 21 | 70 | 25 | 5 | 1.05 | 144.3 | 6.8 | 192.84 | 9.8 |
| 22 | 70 | 25 | 7 | 1.05 | 56.22 | 0.1 | 54.71 | 0.2 |
| 23 | 70 | 25 | 6 | 0.1 | 84.51 | 0.7 | 74.22 | 1.3 |
| 24 | 70 | 25 | 6 | 2 | 170.97 | 17.4 | 214.43 | 24.1 |
| $25^{\text {c }}$ | 70 | 25 | 6 | 1.05 | 181.27 | 6.9 | 238.89 | 9.4 |
| $26^{\text {c }}$ | 70 | 25 | 6 | 1.05 | 155.53 | 5.3 | 101.54 | 7.5 |
| $27^{\text {c }}$ | 70 | 25 | 6 | 1.05 | 205.57 | 0 | 181.00 | 0.0 |

${ }^{\text {c. }}$ centre points.
RSD \%: residual standard deviation percentage.

Regression analysis was carried out to fit mathematical models to the experimental data. The regression coefficients and analysis of variance of the coded independent variables are presented in Table 4.2.

Table 4.2 Regression coefficients and analysis of variance of coded dependent variables.

| Regression coefficients | Isoamyl acetate ( $\mathbf{Y}_{1}$ ) | Furfuryl acetate ( $\mathbf{Y}_{\mathbf{2}}$ ) |
| :---: | :---: | :---: |
| $\beta_{0}$ | 154.78*** | 157.00 |
| $\beta_{1}$ | 79.85 *** | 6.96 |
| $\beta_{2}$ | -30.53* | -7.02 |
| $\beta_{3}$ | -14.15 | -34.93 |
| $\beta_{4}$ | 36.34** | 27.01 |
| $\beta 1^{2}$ | 14.19 | -3.93 |
| $\beta 2^{2}$ | 54.23 | 61.08 |
| $\beta 3^{2}$ | -41.52 | -24.81 |
| $\beta 4^{2}$ | -14.03 | -4.26 |
| $\beta_{12}$ | -15.83 | 12.96 |
| $\beta_{13}$ | 20.81 | 8.52 |
| $\beta_{14}$ | 27.27* | 15.45 |
| $\beta_{23}$ | -8.410 | -21.50 |
| $\beta_{24}$ | 19.91 | 32.62 |
| $\beta_{34}$ | 15.34 | -18.50 |
| $R^{2}$ | 0.90 | 0.44 |
| Regression ( $P$-value) | 0.00*** | 0.768 |
| Lack of fit ( $P$-value) | 0.251 | 0.36 |

[^1]The resulting regression coefficients for the coded dependent variables showed that the regression model for isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ had a statistically good fit. The $R^{2}$ statistic indicated that the response surface model accounted for $90 \%$ of variation in the isoamyl acetate response $\left(\mathrm{Y}_{1}\right)$, which is above the minimum recommended $R^{2}$ value of 0.80 (Joglekar \& May, 1987). For the furfuryl acetate response $\left(\mathrm{Y}_{2}\right)$, only $44 \%$ of the variation could be explained by the response surface model, therefore it had a statistically poor fit and poor prediction capabilities. A very highly significant regression $P$-value ( $\mathrm{p} \leq 0.001$ ) and a non-significant lack of fit $P$-value ( $\mathrm{p} \geq 0.05$ ) indicated that the mathematical model fitted well to the experimental data for isoamyl acetate ( $\mathrm{Y}_{1}$ ) (Bezerra et al., 2008). Although the lack of fit $P$-value was non-significant ( $\mathrm{p} \geq 0.05$ ) for furfuryl acetate response $\left(\mathrm{Y}_{2}\right)$, the regression $P$-value was also nonsignificant ( $\mathrm{p} \geq 0.05$ ). This indicated that the mathematical model did not fit well to the experimental data for furfuryl acetate. Therefore, the prediction of experimental data was not accurate, consequently resulting in no significant effects ( $\mathrm{p}>0.05$ ) of the parameters on the furfuryl acetate response being observed.

The repeatability of the experimental procedure was determined to check the precision of the method by calculating the relative standard deviation percentage (RSD \%) of three replicates for each experimental condition. The RSD \% for isoamyl acetate ranged from 0.02 \% to $21.5 \%$, with a RSD \% mean of $6.3 \%$. The RSD \% for the furfuryl acetate ranged from 0.02 \% to $35.93 \%$, with a RSD \% mean of $10.5 \%$. One possible explanation for this difference in reproducibility and fitting of the model to the experimental data between the responses may be a result of the concentration range of the furfuryl acetate ( $20-30 \mathrm{ppm}$ ) being relatively narrow in comparison to isoamyl acetate ( $50-90 \mathrm{ppm}$ ). Justification of concentration ranges was based on the Flavour and Extract Manufacturers Association (FEMA) reported uses in literature (Burdock,
2010). In addition, preliminary sensory analysis $(\mathrm{n}=12)$ was conducted, during which panellists successfully ranked emulsions containing various concentrations of isoamyl acetate and furfuryl acetate in order of concentration.

Besides showing the optimal conditions for isoamyl acetate emission, the mathematical model identified and described significant effects of the independent variables and some interesting interactions between the variables on the isoamyl acetate response. These interactions are substantial and a 'one variable at a time' (OVAT) approach may have proven too complicated for optimisation.

### 4.3 Effect of independent variables on the responses

The main effects of variables and their significance on the release of isoamyl acetate from the emulsion into the headspace are shown in Figure 4.1. This plot provides a quantitative display of relative importance of the different regression coefficients, and an indication of a positive or negative influence a variable has on the response. The regression coefficients and analysis of variance of dependent variables and their level of significance on the release of both responses are displayed in Table 4.2. The regression results demonstrate that isoamyl acetate concentration ( $\mathrm{x}_{1}$ ) had a very highly significant linear effect ( $\mathrm{p} \leq 0.001$ ) on the headspace release of isoamyl acetate $\left(\mathrm{Y}_{1}\right)$. The influence of this variable on the response proved to be positive, and was found to be the most influential variable affecting isoamyl acetate headspace release (Figure 4.1). This linear relationship can be seen in Figure 4.2, which implies that as isoamyl acetate concentration increases the release of isoamyl acetate from the emulsion into the headspace proportionally increases.


Figure 4.1 Main effects and significant parameters on the recovery of isoamyl acetate from emulsion headspace.

* significant ( $\mathrm{p} \leq 0.05$ ).
** highly significant ( $\mathrm{p} \leq 0.01$ )
*** very highly significant ( $\mathrm{p} \leq 0.001$ )
Error bars display the confidence interval levels at $95 \%$.


Figure 4.2 Main effects plot of isoamyl acetate concentration on isoamyl acetate response.

The regression results demonstrate that furfuryl acetate concentration ( $\mathrm{x}_{2}$ ) had a significant effect ( $\mathrm{p} \leq 0.05$ ) on the isoamyl acetate response $\left(\mathrm{Y}_{1}\right)$ (Table 4.2). Moreover, this effect of furfuryl acetate on the release of isoamyl acetate in to the headspace of the emulsion proved to be a negative effect (Figure 4.1). Hence, increasing furfural acetate concentration in the emulsion resulted in a reduction in the headspace release of isoamyl acetate. This effect could be a result of competition between the volatiles for space on the SPME fibre. It was found in previous studies (Howard et al., 2005; Murray, 2001; Matich et al., 1996), that volatile compounds exhibited competition for extraction sites on the SPME fibres. Higher molecular weight compounds can have the tendency to displace those with lower molecular weights, thereby causing inaccuracies in the relative amounts of analytes present, especially with fibres coated with PDMS. This may explain why an increase in concentration of the higher molecular weight compound
furfuryl acetate resulted in a decrease in the lower molecular weight compound isoamyl acetate being absorbed onto the SPME fibre. A possible solution to this may involve reducing the concentrations of both volatiles to a level that may not result in competition for space on the fibre between the volatiles.

According to the results, salt concentration (x4) was found to have a highly significantly positive linear effect ( $\mathrm{p} \leq 0.01$ ) on the volatile release of isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ (Table 4.2). Thus, an increase in salt led to an increase in overall extraction yield of isoamyl acetate from the emulsion (Figures $4.2 \& 4.3$ ). It would be expected that the addition of salt to the emulsion caused a salting out effect (Rocha et al., 2001).


Figure 4.3 Response surface plot for the effect of salt concentration (\%) on isoamyl acetate response.

Similar observations were also reported in previous studies (Cheong et al., 2011; Cheong et al., 2010; Mirhosseini et al., 2007), where extraction efficiency increased with addition of salt. Cheong et al. (2011) reported that maximum salt concentration (30 $\% \mathrm{w} / \mathrm{v}$ ) resulted in optimal conditions for the extraction of volatiles from a Malaysian soursop pulp sample. Similarly, Mirhosseini et al. (2007) observed that average total peak areas of 121 volatiles in orange beverage emulsions improved by $47 \%$ to $78 \%$ when salt was added to the oil in water emulsions. The salting out phenomena was found to be applicable to more complex foods like vegetable soups, a salt content of 52 \% resulted in an increase of overall concentrations of volatiles into the headspace (Mitchell et al., 2011). These studies investigated the salting out effect in foods at relatively high levels ( $0-60 \%$ ), with a goal for extracting volatiles qualitatively from a purely technical point of view. In the present study, the model system was based around common food emulsions, such as cream sauces, ice creams and dressing. Therefore, a salt concentration range closer to that of the typical level in food products of 0.1-2.0 \% was investigated.

No significant effects ( $\mathrm{p}>0.05$ ) were found for furfuryl acetate concentration on the release of furfuryl acetate $\left(\mathrm{Y}_{2}\right)$ from the emulsion. Additionally, the contour plots for furfuryl acetate release (Figure 4.4) shows a 'saddle point', which is a stationary point of neither a maximum nor minimum response (Myers et al., 2009). This indicates that the optimum conditions for furfuryl acetate release from the emulsion lie outside the experimental range investigated in this study. In addition, the $\mathrm{pH}\left(\mathrm{x}_{3}\right)$ proved to have no significant influence ( $\mathrm{p}>0.05$ ) on isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ or furfuryl acetate (Y2) release from the emulsion. This may be due to the narrow experimental pH range of 5 7, which was selected to fit in with the context of the work being undertaken. Food products such as ice creams and sauces, would be expected to have pH levels within
ranges that would be considered tolerable for human consumption. Similar to these findings, Mirhosseini et al. (2007) found that changing the pH using citric acid and sodium hydrogen carbonate solutions in the range of $\mathrm{pH} 2.5-9.5$ did not lead to a significant effect ( $\mathrm{p}>0.05$ ) on the extraction efficiency of the headspace volatiles of orange beverage emulsion using headspace analysis.


Figure 4.4 Contour plot of furfuryl acetate release from emulsion as a function of furfuryl acetate concentration and pH .

### 4.4 Validation of Optimal Conditions

The optimal conditions for the targeted responses were generated by the Modde 5.0 software (Table 4.3). At optimal conditions (isoamyl acetate $90 \mathrm{ppm}\left(\mathrm{x}_{1}\right)$, furfuryl acetate $30 \mathrm{ppm}\left(\mathrm{x}_{2}\right)$, salt $2 \% \mathrm{w} / \mathrm{v}\left(\mathrm{x}_{3}\right)$ and $\left.\mathrm{pH} 6.02\left(\mathrm{x}_{4}\right)\right)$, the predicted values were 326.41 ppm and 203.65 ppm for isoamyl acetate $\left(\mathrm{Y}_{1}\right)$ and furfuryl acetate $\left(\mathrm{Y}_{2}\right)$ release
from the emulsion respectively. HS-SPME GCMS analysis was carried out at the optimal conditions to verify the model. At optimal conditions, the product contained isoamyl acetate (273.75 ppm) ( $\mathrm{Y}_{1}$ ) and furfuryl acetate $(148.40 \mathrm{ppm})\left(\mathrm{Y}_{2}\right)$, which was within the error ranges (241.57-404.77 ppm for isoamyl acetate and $85.42-334.86$ ppm for furfuryl acetate).

Table 4.3 List of predicted conditions generated by Modde 5.0 RSM software.

| Isoamyl <br> acetate <br> $(\mathbf{p p m})\left(\mathbf{x}_{\mathbf{1}}\right)$ | Furfuryl <br> acetate <br> $(\mathbf{p p m})\left(\mathbf{x}_{\mathbf{2}}\right)$ | $\mathbf{p H}$ <br> $(\mathbf{x} \mathbf{3})$ | Salt <br> $(\mathbf{w} / \mathbf{v} \mathbf{\%})$ <br> $(\mathbf{x} \mathbf{4})$ | Isoamyl <br> acetate <br> response $\left(\mathbf{Y}_{\mathbf{1}}\right)$ | Furfuryl <br> acetate <br> response $\left(\mathbf{Y}_{\mathbf{2}}\right)$ |
| :---: | :---: | :---: | :---: | :---: | :---: |
| 90 | 20 | 5.76 | 2 | 369.44 | 153.76 |
| 90 | 20 | 5.89 | 2 | 375.26 | 152.45 |
| 90 | 30 | 5.11 | 2 | 280.30 | 229.85 |
| $\mathbf{9 0}^{*}$ | $\mathbf{3 0}$ | $\mathbf{6 . 0 2}$ | $\mathbf{2}$ | $\mathbf{3 2 6 . 1 8}$ | $\mathbf{2 0 3 . 6 5}$ |
| 90 | 25.37 | 5.6 | 2 | 279.55 | 148.37 |
| 90 | 20 | 5.4 | 2 | 345.88 | 154.42 |
| 90 | 30 | 5.5 | 2 | 306.17 | 223.47 |
| 90 | 20 | 5.57 | 2 | 358.28 | 154.58 |

*optimal condition selected by Modde 5.0 software from the list of generated optimal conditions.

The way in which flavour components modify the perceived flavour of food has advanced from a stage of simple awareness to an understanding of the interactions involved at a qualitative and quantitative level (Taylor \& Hort, 2007). From a practical point of view, optimising the release from the food matrix of two flavour compounds that are strongly associated with positive fruity notes in foods can be a method of enhancing flavour perception. The perception of aroma and flavour is dependent upon the concentration and odour threshold of the volatile compounds present in the food
(Guichard, 2002). Thus, finding the optimum concentration of volatile compounds for maximum flavour release is a crucial step in the development of flavoursome food emulsions, which are an important class of food colloids.

### 4.5 Conclusions

The findings of this chapter provide an insight into the role that salt concentration can have on the volatile release of isoamyl acetate from oil in water emulsion based products. Increasing salt content resulted in an increase in headspace release of isoamyl acetate from the emulsion. Thus, the practical application of such knowledge could enable chefs and food manufacturers to optimise conditions for maximum flavour release of volatile compounds which are strongly associated with fruity notes in food emulsion products. The main effects of isoamyl acetate concentration and salt concentration should be considered as critical factors when studying the release of isoamyl acetate from an oil in water emulsion. As previously discussed in section 3.6, there are limitations to application of knowledge obtained from food model systems in terms of their transfer to more complex foods. In terms of the transferability of the findings to an ice cream product, factors such as processing temperatures (below freezing), sugar content and protein content would also influence organic volatile release, and must be considered.

## Chapter 5

### 5.1 General introduction

This chapter involved the investigation of the pairing of food based on perceived flavour as an important sensory phenomenon. The palatability of a food is largely determined by flavour (Breslin \& Beaunchamp, 1995), and for this reason, flavour is one of the most imperative attributes of food in terms of determining consumer acceptance (Liu \& Yang, 2002). In an increasingly globalised food market, the desire for a competitive advantage has resulted in a search for more unique flavour pairings becoming a constant preoccupation of food product developers (de Klepper, 2011). Despite recent advances, the scientific literature on flavour pairing is surprisingly limited, especially given the enormous scientific and commercial progress towards a better understanding of flavour pairing (Møller, 2013). Some studies have focused on the hedonic response to pairing chocolate with a wide range of beverages (teas and alcoholic beverages) (Donadini et al., 2012) and on preferences for wine and cheese pairings (Bastian et al., 2010).

A literature search reveals a lack of research papers published which explore the pairing of foods, only two studies to date were found that explore the impact of volatile compounds on successfully pairing foods. Kort et al. (2010) used a more sensory driven approach, exploring food pairings through sensory evaluation in conjunction with a volatile database. Ahn et al. (2011) explored the impact volatile compounds had on the selection of ingredients using a volatile database and several food ingredient databases. Hence, no studies have explored the phenomena of food pairing using the holistic approach of organic volatile analysis, hedonic response evaluation and descriptive sensory analysis.

The aim of this chapter was to investigate food pairings as an important sensory phenomenon with a key interest in determining how different components in the selected food pairings (both volatile and non-volatile) affect and interact with other components to influence sensory perception. While the food pairings investigated in this chapter can be found in Asian, African and South American countries, they would be considered novel and unique in a European context, and were therefore of interest. The selected ingredients comprise a variety of food categories; fish, meat, starchy food, fruit and lipid. A conjoint approach utilising qualitative (organic volatile analysis, focus groups and descriptive sensory analysis) and quantitative (comparable semi quantitative organic volatile analysis and affective sensory tests) methods of analysis was performed in an attempt to elucidate the success or failure of selected food pairings.

### 5.2 Focus group

### 5.2.1 Participants' attitudes and opinions towards the paring of novel foods

Participants of the focus groups were familiar with the concept of pairing foods together based on flavours. The majority of daily meals or snacks prepared or consumed by the participants were based on pairing foods that complement each other from a flavour perspective. Pairing foods based on flavours was something that participants considered to be learned from experience and/or exposure through individual preferences, and cultural and environmental influences (friends, family and personal experiences) as opposed to being innate. In addition, participants were accustomed to the known/traditional food pairings that were mentioned by the focus group moderators (apple and cinnamon, wine and cheeses and steak and onions, etc.). Participants also discussed other known/traditional personal food pairings (orange and chocolate, mint and chocolate and strawberries and vanilla, etc.).

From the discussions about successfully pairing foods, it can be concluded that combining contrasting foods from different taste categories (sweet, savoury, bitter, salty and sour) is a common and popular method of producing flavoursome food pairings. In particular, combining sweet and sour, bitter and sweet and sweet and salty contrasting tastes were mentioned. This amalgamation of contrasting flavours has been noted as a pattern that manifests itself in East Asian cuisines. North American and Western European dishes tend to combine ingredients that share flavour compounds, while East Asian cuisine avoids those (Ahn et al., 2011). Hence, based on these results, it would appear that there is a certain degree of influence from the recipes of East Asian cuisines on the way meals are constructed by the participants (who were predominantly of Western European origin) in the focus groups.

Many participants had their own personal food pairings which they have tried and tested themselves, for example fried egg with strawberry jam and bread, jam and mustard. Although there was a variety of personal food pairings mentioned, the conclusion reached was that an imperative factor in successfully pairing foods was the method of cooking and presentation of the foods. For example, egg, bread and jam was discussed as not being a very appealing food pairing, however, if presented as French toast (bread soaked in egg and fried) with jam it would be very appealing. A discussion took place about how many of the food pairings of today have evolved from the restaurant environment (as menu items) and gained appeal resulting in the food manufacturing industry producing them as products. It was also made clear that pairing novel flavours was identified as being a marketing tool of many food companies in order to create interest in their products, with some products being very successful (for example cheese and onion crisps). Participants initially liked the aromas of the food pairings during the blind evaluation. However, once all the foods were identified, many
of the participant's attitudes towards them changed from positive to negative, demonstrating a certain degree of neophobic behaviour. Many participants explained that they would have to deliberate whether or not to consume some of the novel food pairings (in particular banana and extra virgin olive oil, banana and bacon and banana and blue cheese).

### 5.2.2 Orthonasal sensory evaluation of food pairings

Mean acceptability scores with standard deviations for evaluation of orthonasal aroma by the focus group panel $(\mathrm{n}=19)$ are displayed in Figure 5.1. A mean acceptability score of five was considered to be a limit of acceptance (Mitchell et al., 2013). The pairings of banana and basmati rice $(B+R)$, banana and bacon $(B+B N)$ and banana and extra virgin olive oil ( $\mathrm{B}+\mathrm{O}$ ) with scores of $5.6 \pm 1.1,5.4 \pm 1.8$ and $6.4 \pm 2.2$ respectively were the only samples above this limit. In addition, no significant differences ( $\mathrm{p}>0.05$ ) in acceptability were found between these three food pairings (Figure 5.1). Conversely, the pairings of banana and mackerel and banana and blue cheese were found to be significantly less acceptable ( $\mathrm{p} \leq 0.05$ ) than all other pairings with banana ( $2.2 \pm 1.5$ and $3.8 \pm 2.3$ respectively).

Following the evaluation of the food pairings an open discussion regarding the samples took place, during which a general consensus for ranking of the food pairings based on preference was conducted and discussed. Banana and basmati rice was established as being consensually the most preferred food pairing. This food pairing was considered to be suitable for both a savoury or sweet food product. It was concluded that banana and bacon was the second preferred food pairing, followed by banana and extra virgin olive oil.


Foods paired with banana
Figure 5.1 Mean acceptability scores for orthonasal aroma evaluation of food pairings.
Each value is presented as a mean $\pm \operatorname{SD}(\mathrm{n}=19)$.
Samples with different letters are significantly different ( $p \leq 0.05$ ).

-     -         - : limit of acceptability.


### 5.3 Consumer sensory evaluation of food pairings

The mean acceptability scores $(\mathrm{n}=85)$ for the three preferred food pairings samples are illustrated in Figure 5.2. No significant differences ( $p>0.05$ ) were found between the acceptability scores of banana and basmati rice $(B+R)$ and banana and bacon $(B+B N)$ which were $6.1 \pm 1.6$ and $6.2 \pm 2.1$ respectively. Banana and extra virgin olive oil ( $\mathrm{B}+\mathrm{O}$ ) received a significantly lower ( $\mathrm{p} \leq 0.05$ ) mean acceptability scoring ( 3.4 $\pm 2.2$ ) in comparison to $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$. The mean acceptability score for $\mathrm{B}+\mathrm{O}$ was below the limit of acceptability for this study $(<5)$, while $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$ were above this limit $(\geq 5)$.


Figure 5.2 Mean scores for hedonic ratings of food pairings with banana.
Each value is presented as a mean $\pm$ SD $(n=85)$.
Samples with different letters are significantly different ( $\mathrm{p} \leq 0.05$ ).

-     -         - : limit of acceptability.

The frequency of positive and negative comments from the consumers as a percentage of total subjects $(\mathrm{n}=85)$ is represented in Table 5.1. The majority of comments regarding $\mathrm{B}+\mathrm{BN}$ and $\mathrm{B}+\mathrm{R}$ were positive ( $68 \%$ and $65 \%$ respectively), such as "the flavours do work well, I didn't expect to like it" and "the two foods complement each other". Conversely, the vast majority of panellists ( $82 \%$ ) provided negative comments for $\mathrm{B}+\mathrm{O}$, such as "the pairing doesn't work" and "the oil is slightly bitter, grassy and overpowers the fruit".

Table 5.1 Consumer comments (positive and negative) represented as a percentage (\%) of total number of subjects $(\mathrm{n}=85)$ for the three food pairings.

| Food pairing sample | Positive comment \% | Negative comment \% |
| :--- | :---: | :---: |
| Banana and basmati rice | 65 | 35 |
| Banana and bacon | 68 | 32 |
| Banana and extra virgin olive oil | 18 | 82 |

The preference test results (Figure 5.3 ) showed that $\mathrm{B}+\mathrm{BN}$ had the highest percentage of $1^{\text {st }}$ preference choices $(55.3 \%), \mathrm{B}+\mathrm{R}$ had the highest percentage of $2^{\text {nd }}$ choice preferences ( $49.4 \%$ ) and $\mathrm{B}+\mathrm{O}$ had the highest percentage of $3^{\text {rd }}$ preference choices $(77.6 \%)$. The cross tabulation results corroborate the results for the hedonic ratings of the samples, where $\mathrm{B}+\mathrm{BN}$ and $\mathrm{B}+\mathrm{R}$ scored significantly higher ( $\mathrm{p} \leq 0.05$ ) than B+O. Additionally, results from the Pearson's Chi-squared were very highly significant ( $\mathrm{p} \leq 0.001$ ), and the Cramer's V coefficient value ( 0.5 ) indicated that there was a high association between the sample type and preference. Based on the quantitative and qualitative analysis, it can be concluded that banana and basmati rice $(B+R)$ and banana and bacon ( $B+B N$ ) were considered to be acceptable food pairings. What's more, $\mathrm{B}+\mathrm{BN}$ was preferred the most, while $\mathrm{B}+\mathrm{R}$ was the second most preferred food pairing. Banana and extra virgin olive oil $(\mathrm{B}+\mathrm{O})$ was deemed to be the least preferred and an unacceptable food pairing.


Figure 5.3 Percentage of choice of $1^{\text {st }}, 2^{\text {nd }}$ and $3^{\text {rd }}$ preference of food pairings from the consumer panel $(\mathrm{n}=85)$.

### 5.4 Descriptive analysis

The orthonasal (odour) and retronasal (flavour) aroma descriptors for the three food pairings along with their frequency of occurrence represented as a percentage of total subjects $(\mathrm{n}=28)$ are shown in Table 5.2. It can be seen that the majority of assessors (>50\%) described the odour and flavour of banana and basmati rice ( $\mathrm{B}+\mathrm{R}$ ) as sweet and cereal. The odour and flavour of banana and bacon (B+BN) was described as sweet, caramel, salty, fried meat and meaty (>50\%). While, the odour and flavour of banana and extra virgin olive oil ( $\mathrm{B}+\mathrm{O}$ ) was described as bitter, chemical and fatty/oily (> $50 \%$ ).

Table 5.2 List of orthonasal (A) and retronasal (F) aroma sensory descriptive attributes and their frequency represented as a percentage total (\%) number of subjects $(\mathrm{n}=28)$ generated by Free Choice Profiling ( FCP ) for the three food pairings.

|  | B+R |  | B+BN |  | B+O |  |  | B+R |  | B+BN |  | B+O |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | A | F | A | F | A | F |  | A | F | A | F | A | F |
| Sweet |  |  |  |  |  |  | Empyreumatic |  |  |  |  |  |  |
| Sweet | 62 | 55 | 69 | 97 | 48 | 41 | Smoky |  |  | 38 | 24 |  |  |
| Caramel |  |  | 62 | 48 |  |  | Woody | 24 | 7 |  |  |  | 21 |
| Maple |  |  |  | 28 |  |  | fried meat |  |  | 66 | 48 |  |  |
| Starchy |  |  |  |  |  |  | Floral |  |  |  |  |  |  |
| Cereal | 62 | 66 |  |  |  |  | Floral | 45 | 10 | 25 | 21 | 38 | 24 |
| Starchy | 41 | 14 | 31 | 14 |  |  | Herbal |  |  |  |  | 38 |  |
| Popcorn | 28 | 31 |  |  |  |  | Olive |  |  |  |  | 34 | 55 |
| Fruit |  |  |  |  |  |  | Fat |  |  |  |  |  |  |
| Fruity |  | 45 | 31 | 59 | 31 | 28 | Fatty/Oily |  |  | 34 | 52 | 55 | 83 |
| Pear/Apple | 7 | 3 |  |  |  |  | Buttery | 10 |  | 10 |  |  |  |
| Misc. |  |  |  |  |  |  | Savoury |  |  |  |  |  |  |
| Citric |  |  |  |  |  | 10 | Meaty |  |  | 66 | 86 |  |  |
| Salty |  |  | 52 | 72 |  |  | Savoury |  |  | 41 | 38 |  |  |
| Bitter | 10 | 14 | 3 | 3 | 52 | 72 | Umami |  |  |  | 28 |  |  |
| Chemical/Soapy | 3 |  |  |  | 72 | 66 | Earthy |  |  |  |  |  |  |
| Acidic |  |  | 3 | 3 | 31 | 38 | Earthy | 48 | 28 | 7 |  | 17 | 17 |
| Watery | 28 | 41 |  |  |  |  | Pine |  |  |  |  | 17 | 3 |
| Spicy/Peppery |  |  |  |  | 45 | 41 | Nutty | 38 | 34 |  | 24 | 21 | 14 |
|  |  |  |  |  |  |  | Grassy | 21 | 10 | 7 | 3 | 28 | 28 |

Italics indicates orthonasal aroma and retronasal aroma (flavour) classification obtained from Fisher \& Scott, 1997.
A: indicates orthonasal aroma (odour) descriptive term. F: indicates retronasal aroma (flavour) descriptive term.

A consensus biplot of the orthonasal and retronasal descriptive terms and their correlation with the unpaired and paired samples generated by the General Procrustes Analysis (GPA) is presented in Figure 5.4. From this plot it can be seen that the food pairings were separated into different quadrants through the correlated consensual sensory descriptors for orthonasal and retronasal aroma from factor 1 and factor 2 . The correlations between the orthonasal and retronasal sensory attributes and the first two factors of the GPA analysis are presented in Table 5.3. The higher correlation levels ( $\geq 0.8$ and $\leq-0.8$ ) indicate that factor 1 was more associated with oily orthonasal (bitter, soapy/chemical, herbal, olive, spicy/peppery and pine-aromas) and retronasal (smoky, buttery, floral and olive flavours) sensory properties. While, factor 2 appeared to be more associated with meaty orthonasal (earthy, meaty, fried meat, savoury, salty, smoky and fatty/oily aromas) and retronasal (earthy, meaty, fried meat, savoury, salty, umami and fatty/oily flavours) sensory properties. It seems, the assessors perceived the difference between the formulations in this study as being based on meaty and oily properties, which allowed them to preferentially choose certain food pairings.


Figure 5.4 Consensus biplot for orthonasal (A) and retronasal (F) aroma sensory descriptors and samples generated by Generalised Procrustes Analysis.

* : food samples (unpaired and paired).
- descriptive sensory terms for orthonasal (A) and retronasal (F) aroma generated by FCP.

Table 5.3 Correlation coefficients for General Procrustes Analysis of the first two factors.

| Orthonasal terms | Factor 1 | Factor 2 | Retronasal terms | Factor 1 | Factor 2 |
| :--- | :---: | :---: | :--- | :---: | :---: |
| Earthy-A | -0.014 | -0.829 | Earthy-F | -0.014 | -0.829 |
| Bitter-A | 0.970 | 0.118 | Floral-F | 0.970 | 0.118 |
| Soapy/Chemical-A | 0.925 | 0.078 | Caramel-F | 0.925 | 0.078 |
| Herbal-A | 0.972 | 0.202 | Nutty-F | 0.972 | 0.202 |
| Fatty/Oily-A | 0.499 | 0.847 | Fatty/oily-F | 0.499 | 0.847 |
| Olive-A | 0.849 | 0.128 | Olive-F | 0.849 | 0.128 |
| Spicy/Peppery-A | 0.952 | 0.209 | Buttery-F | 0.952 | 0.209 |
| Pine-A | 0.857 | 0.130 | Smoky-F | 0.857 | 0.130 |
| Meaty-A | -0.554 | 0.816 | Meaty-F | -0.554 | 0.816 |
| Savoury-A | -0.512 | 0.833 | Fried meat-F | -0.512 | 0.833 |
| Salty-A | -0.535 | 0.814 | Salty-F | -0.535 | 0.814 |
| Smoky-A | -0.551 | 0.814 | Savoury-F | -0.551 | 0.814 |
| Fried meat-A | -0.564 | 0.809 | Umami-F | -0.564 | 0.809 |

A: orthonasal descriptive term. F: retronasal descriptive term.

It can also be seen from Figure 5.4 that the food pairings of $B+R$ and $B+B N$ directionally correlated with their respective unpaired food ingredients (basmati rice for $\mathrm{B}+\mathrm{R}$ and bacon for $\mathrm{B}+\mathrm{BN}$ ). Additionally, the characteristic descriptive flavour and aroma terms for unpaired bacon, basmati rice and extra virgin olive oil showed directional correlation with their respective unpaired foods. In the top left hand side of the plot, flavour and odour descriptive terms of meaty, fried meat, smoky and salty covary with bacon, and have been previously associated with bacon flavour and aroma (Kathrine et al., 2013; Timón et al., 2004; Maw et al., 2001). Similarly, previously associated sensory descriptive terms for rice (popcorn, starchy, earthy and sweet) covary with basmati rice in the lower left hand side of the plot (Bryant \& McClung, 2011; Limpawattana et al. 2008; Bhattacharjee et al., 2002).

The associated characteristic sensory descriptive terms for extra virgin olive oil (fatty/ oily, soapy/ chemical, grassy, bitter, citrus and nutty) also covary with extra virgin olive oil (Tanouti et al., 2012; Delgado \& Guinard, 2011). The significant estimated regression coefficients for the correlation between the unpaired and paired samples and the orthonasal and retronasal aroma descriptive terms generated during FCP analysed by APLSR are shown in Table 5.4. Numerous significant positive effects ( $\mathrm{p} \leq 0.05$ ) between descriptive terms and food samples were found. B+BN was found to have a highly significant correlation to meaty aroma ( $\mathrm{p} \leq 0.01$ ) and significant correlation to meaty flavour ( $\mathrm{p} \leq 0.05$ ).

Table 5.4 Significance of estimated regression coefficients (ANOVA values) for the relationships between the samples (unpaired and paired) and the orthonasal (A) and retronasal ( F ) aroma sensory attributes.

| Descriptors | Banana | Oil | Bacon | Rice | $\mathbf{B}+\mathbf{R}$ | $\mathbf{B}+\mathbf{B N}$ | $\mathbf{B + O}$ |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| Sweet-A | $0.18^{* *}$ | -0.17 | -0.04 | -0.03 | 0.08 | 0.00 | -0.03 |
| Meaty-A | -0.84 | 0.79 | 0.18 | 0.12 | -0.37 | $0.002^{* *}$ | 0.12 |
| Acidic-F | $-0.19^{*}$ | 0.18 | 0.04 | 0.03 | -0.09 | 0.00 | 0.03 |
| Woody-F | $-0.19^{*}$ | 0.18 | 0.04 | 0.03 | -0.08 | 0.00 | 0.03 |
| Bitter-F | $-0.24^{* * *}$ | 0.23 | 0.05 | 0.04 | -0.11 | 0.00 | 0.03 |
| Fatty/oily-F | -0.22 | $0.20^{* *}$ | 0.05 | 0.03 | -0.10 | 0.00 | 0.03 |
| Meaty-F | -0.54 | 0.50 | 0.11 | 0.08 | -0.24 | $0.001^{*}$ | 0.08 |

ANOVA APLSR values, the sign dictates whether the correlation is positively or negatively correlated.

* significant ( $\mathrm{p} \leq 0.05$ ).
** highly significant ( $\mathrm{p} \leq 0.01$ ).
*** very highly significant $(\mathrm{p} \leq 0.001)$
A: aroma descriptive term.
F: flavour descriptive term.


### 5.5 Organic volatile compound profile analysis

In total, 119 organic volatile compounds were detected in the samples (see appendix E for full list). Esters were identified as being most abundant, with 50 ester volatiles detected in total. Alcohols were the second most abundant group of volatiles detected, with 23 detected. Twenty one aldehydes and 10 ketones were also detected. Other compounds that were detected in lesser quantities were; sulphurs (four), phenyls (three), alkenes (three), pyrazines (two), carboxylic acids (two), terpenes (one), phenols (one) and furans (one). From the 119 organic volatile compounds detected in the samples, only four of the volatile compounds (3-pentanone and 2, 4-heptadienal (both absent from $\mathrm{B}+\mathrm{BN}$ ), ethyl hexanoate (absent from $\mathrm{B}+\mathrm{R}$ ) and amyl isovalerate (absent from $\mathrm{B}+\mathrm{O})$ ) were not present in the headspace of all the food pairings.

The headspace of the banana was found to contain the most abundant number of organic volatile compounds with 118 being detected, followed by the extra virgin olive oil (102 organic volatile compounds), bacon ( 98 organic volatile compounds) and finally basmati rice (86 organic volatile compounds). In addition, 11 organic volatile compounds (mainly esters) were detected only in banana and not in the other unpaired samples (Table 5.5). The significant ANOVA values for the organic volatile compounds which were detected in the samples and that are considered to contribute to the characteristic aroma and flavour of their corresponding foods as suggested by the literature are presented in Table 5.6. Numerous significant correlations were found between the organic volatile compounds and the unpaired and paired samples. These results corroborate the orthonasal and retronasal sensory attribute regression coefficient values, where banana was found to have a highly significant positive correlation ( $\mathrm{p} \leq$ 0.01 ) with sweet aroma (Table 5.4).

Table 5.5 List of organic volatile compounds detected in the food pairings which were contributed by banana only.

| Volatile compound | $\mathbf{B}$ | $\mathbf{O}$ | $\mathbf{R}$ | $\mathbf{B N}$ | $\mathbf{B + B N}$ | $\mathbf{B +}+\mathbf{R}$ | $\mathbf{B + O}$ | $\mathbf{R T}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| acetyl valeryl | P | NP | NP | NP | P | P | P | 16.8 |
| 2-methylbutyl propanoate | P | NP | NP | NP | P | P | P | 21.7 |
| 1-methylbutyl butanoate | P | NP | NP | NP | P | P | P | 24.6 |
| 2-methylbutyl butanoate | P | NP | NP | NP | P | P | P | 25.3 |
| isoamyl butyrate | P | NP | NP | NP | P | P | P | 25.6 |
| 3-methyl, 6-hepten-1-ol | P | NP | NP | NP | P | P | P | 26.5 |
| amyl isovalerate | P | NP | NP | NP | P | P | NP | 26.6 |
| butyl pentanoate | P | NP | NP | NP | P | P | P | 27.1 |
| 1-methyl, octyl butanoate | P | NP | NP | NP | P | P | P | 27.7 |
| 3-octen-1-ol acetate | P | NP | NP | NP | P | P | P | 29.2 |
| hexyl 2-methylbutanoate | P | NP | NP | NP | P | P | P | 29.8 |
| hexyl 3-methylbutanoate | P | NP | NP | NP | P | P | P | 29.8 |

P: compound present in sample, NP: compound not present in sample.
RT: retention time in minutes.
Unpaired samples: B: Banana, O: extra virgin olive oil, BN: bacon, R: basmati rice.
Paired samples: $\mathrm{B}+\mathrm{R}$ : banana and basmati rice, $\mathrm{B}+\mathrm{BN}$ : banana and bacon, $\mathrm{B}+\mathrm{O}$ banana and extra virgin olive oil.

From the organic volatile compounds analysis results it would seem that banana contributed greatly to the volatile profiles of the food pairings (Table 5.5 and Figure 5.5). Amyl and butyl esters have been previously identified as being the most abundant and the most odour active compounds in fresh banana eliciting the characteristic fruity banana notes (Imahori et al., 2013; Pino \& Felbes, 2013; Vermeir et al., 2009; Boudhrioua et al., 2003; Mayr et al., 2003; Liu \& Yang, 2002). Of these esters, acetates are of particular importance due to their high concentrations and low odour thresholds (Pontes et al., 2012). In particular, isoamyl acetate, pentyl acetate, isoamyl butyrate, 3methylbutyl 3-methylbutanoate, 3-methylbutyl 2-methylbutanoate and isoamyl isovalerate which were found (Table 5.6) to have a very highly significant positive correlation with banana ( $\mathrm{p} \leq 0.001$ ).

Table 5.6 Significance of estimated regression coefficients (ANOVA values) for the relationships between samples (unpaired and paired) and volatile compounds detected by SPME GCMS.

| RT | Volatile compounds | B | O | BN | R | B+R | B+BN | B+O |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 6.5 | ethanol | -0.24*** | 0.22*** | 0.05** | 0.03*** | -0.11* | 0.00** | 0.03* |
| 9 | ethyl acetate | -0.38* | 0.35*** | 0.08 ns | $0.05 * * *$ | -0.17 ns | 0.01 ns | 0.05* |
| 9.2 | acetic acid | -0.41** | 0.38*** | 0.09** | 0.06 ns | $-0.18^{* * *}$ | 0.01*** | 0.06* |
| 14 | 1-pentanol | -0.40** | 0.38*** | 0.09 ns | 0.06*** | -0.18* | 0.01*** | 0.06* |
| 18 | isoamyl acetate | 0.55*** | $-0.51 * * *$ | -0.12** | -0.08*** | 0.25* | -0.01 ns | -0.07 ns |
| 18 | p-xylene | -0.48*** | $0.44 * * *$ | 0.10 ns | 0.07** | -0.21 *** | 0.01* | 0.07* |
| 20 | pentyl acetate | 0.50*** | -0.46*** | -0.11*** | -0.07* | $0.22^{* * *}$ | $-0.01 * * *$ | -0.07*** |
| 20 | heptanal | $-0.27 * * *$ | 0.25*** | 0.06* | 0.04*** | -0.12 ns | 0.01** | 0.04*** |
| 21 | pinene | -0.41** | 0.38*** | 0.09 ns | 0.06** | -0.18 ns | 0.01** | 0.06*** |
| 22 | 2-heptenal | -0.45*** | $0.42^{* * *}$ | 0.10 | 0.06*** | -0.20*** | 0.01*** | 0.06** |
| 22 | benzaldehyde | -0.01 ns | 0.01*** | 0.00 | 0.00*** | -0.01 ns | 0.00** | 0.00** |
| 24 | octanal | -0.20** | 0.19 ns | 0.04* | 0.03*** | -0.09 ns | 0.00*** | 0.03* |
| 26 | isoamyl butyrate | 0.57*** | $-0.53^{* * *}$ | -0.12** | $-0.08 * * *$ | 0.25* | $-0.01 * * *$ | -0.08** |
| 27 | 3-methylbutyl 2methylbutanoate | 0.55*** | $-0.52^{* * *}$ | -0.12** | -0.08*** | 0.25* | -0.01* | $-0.08^{* * *}$ |
| 27 | 3-methylbutyl 3methylbutanoate | 0.51*** | -0.48*** | -0.11*** | -0.07*** | 0.23** | -0.01*** | -0.07*** |
| 27 | isoamyl isovalerate | 0.51*** | -0.48 ns | -0.11** | -0.07*** | 0.23* | -0.01*** | -0.07 *** |
| 29 | decanal | $-0.41 \mathrm{~ns}$ | $0.38^{* * *}$ | 0.09* | 0.06*** | -0.18** | 0.01** | 0.06 ns |

ANOVA APLSR values, the sign dictates whether the correlation is positively or negatively correlated.

* significant ( $\mathrm{p} \leq 0.05$ ).
** highly significant ( $\mathrm{p} \leq 0.01$ ).
*** very highly significant $(\mathrm{p} \leq 0.001)$
RT: retention time.
Unpaired samples: B: Banana, O: extra virgin olive oil, BN: bacon, R: basmati rice.
Paired samples: $\mathrm{B}+\mathrm{R}$ : banana and basmati rice, $\mathrm{B}+\mathrm{BN}$ : banana and bacon, $\mathrm{B}+\mathrm{O}$ banana and extra virgin olive oil.


Figure 5.5 Chromatogram of unpaired banana sample and paired food samples.
a: banana and bacon, b : banana and extra virgin olive oil, c : banana and basmati rice, d : unpaired banana.

What's more, the volatile compounds mentioned above that correlated with banana were also found to have very highly significant positive correlations ( $\mathrm{p} \leq 0.001$ ) with $B+R$ while having either no significant correlations ( $p>0.05$ ) or very highly significant negative correlations ( $\mathrm{p} \leq 0.001$ ) with the other food pairings ( $\mathrm{B}+\mathrm{O}$ and $B+B N$ ). These results corroborate the descriptive sensory results, where $B+R$ and banana covaried in the biplot (Figure 5.4).

An apparent change in the distribution coefficient appeared to occur at an approximate retention time of 18 minutes onwards, in which the release of the more non polar (lipophilic) compounds (mainly esters) appeared to be suppressed (Figure 5.5). This suppression in lipophilic compounds seemed to be more prominent in $B+B N$ and $B+O$, than in $B+R$. Such a phenomenon is more than likely due to the binding of these lipophilic compounds to the food matrices. It is well known that food matrix components can bind, entrap or encapsulate volatile flavour compounds, resulting in a reduction in the rate of flavour release and flavour intensity (Naknean \& Meenune, 2010). Lipid content and the volatile compounds lipophilicity are known key factors affecting flavour release in foods (Philippe et al., 2006).

The effect of lipids on flavour release and perception is complex, and affects both the release of stimuli from the food matrix and also the release during oral processing of foods (Arancibia et al., 2011). For instance, flavour release of lipophilic aroma compounds was reported to decrease with increasing lipid levels in the food matrix (Linforth et al., 2010). Thus, a strong affinity of more lipophilic volatile compounds for the lipid content in both $\mathrm{B}+\mathrm{BN}$ and in $\mathrm{B}+\mathrm{O}$ may have resulted in a retention of such volatiles, thereby influencing the flavour profile and influencing the perceived sensory perception. As expected, the low fat content of the rice in $\mathrm{B}+\mathrm{R}$ did not alter the fruit flavour eliciting compounds.

### 5.6 Volatile interactions

The individual foods (unpaired) of banana, basmati rice, bacon and extra virgin olive oil share the majority of volatile compounds, therefore possible synergistic and/or antagonistic volatile-volatile interactions occurred resulting in positive or negative hedonic responses from the assessors. When discussing combining flavours, odour intensity (compound concentration in the foods and flavour thresholds) must be taken into account (Ahn et al., 2011). A possible volatilevolatile interaction regarding odour intensity may have occurred. An increase in the concentrations of certain volatile compounds (with low initial concentrations) to a level that exceeds their odour threshold would result in these volatile compounds having more of a contribution to the flavour profile and thereby altering it (Belitz et al., 2009). In the case of $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$, these interactions were positive as is suggested by the hedonic results (Figures 5.2 \& 5.3 and Table 5.1 ), whereas with B+O, the interaction was a negative. Furthermore, the perceived aroma of certain compounds can be altered as concentrations change. It has been found that hexanal and nonanal in extra virgin olive oil elicit herb olive odours at low concentrations, changing to rancid odours at higher concentrations (Aparicio et al., 2000). Therefore in the case of B+O, compounds which may have initially elicited pleasant aromas in the extra virgin olive oil may have contributed negatively to the perceived flavour profile of food pairing once paired with banana due to a cumulative increase in concentration of such compounds.

Another possible volatile-volatile interaction may have involved the odour active compounds of the constituents of the food pairings. In the case of $B+R$ (esters from banana and aldehydes from basmati rice) and B+BN (esters from
banana and sulphurous and pyrazines from bacon) the pairing of these foods produced harmonious and pleasant volatile mixtures from a sensory perspective (Figure 5.2 and Table 5.1). Whereas, in the case of $\mathrm{B}+\mathrm{O}$, the combination of odour active compounds in extra virgin olive oil (mainly aldehydes, ketones and alkenes) and banana appeared to produce an unpleasantly perceived volatile mixture.

Creating a balanced flavour mixture is another important aspect to take into consideration when discussing pairing of foods. As the concentration of sharing volatiles increases, subsequently the volatile concentration ratio can be altered in such a way that certain compounds predominate the flavour profile (Belitz et al., 2009). It appears that the pleasant banana eliciting compounds dominated the flavour profile of $\mathrm{B}+\mathrm{R}$ more so than in $\mathrm{B}+\mathrm{BN}$ and $\mathrm{B}+\mathrm{O}$. This is clear in Figure 5.4 , as $\mathrm{B}+\mathrm{R}$ covaried with banana, while being directionally correlated with descriptive terms of fruity, pear/apple, sweet aromas and flavours. In contrast to this, extra virgin olive oil appears to dominate the flavour profile of $\mathrm{B}+\mathrm{O}$, as $\mathrm{B}+\mathrm{O}$ is positioned closer to the extra virgin olive oil and its associated descriptive terms (spicy, soapy/chemical, bitter and oily/fatty) in the biplot (Tanouti et al., 2012; Angerosa et al., 2000). Moreover, pinene which has previously been identified as the active compound for the hot spicy flavour in virgin olive oils (Ekundayo et al, 1988), was found to have a significant positive correlation ( $\mathrm{p} \leq 0.001$ ) with oil and $\mathrm{B}+\mathrm{O}$ (Table 5.6), suggesting that the lipid content caused an increase in the hot spicy flavour perceived in $\mathrm{B}+\mathrm{O}$. In addition, acetic acid, ethanol and ethyl acetate which have all previously been associated with vinegar or wine flavours in olive oils (Kalua et al., 2007), were found to have significant positive correlations with $\mathrm{B}+\mathrm{O}(\mathrm{p} \leq 0.05)$.

Although the foods that were investigated were found to share the majority of volatile compounds, it would be extremely difficult to support the hypothesis that the success of the food pairings is based on that fact alone. Although banana and extra virgin olive oil was found to be an unsuccessful food pairing, it must be noted that the original food pairing suggested by foodpairing.com was not investigated. This software suggested that the food pairing of banana and an extra virgin olive oil produced from a specific variety of olive ('Family Reserve Arbequina'), the variety of grape used in the production of the oil used in this study is not specified by the manufacturer. Hence this exact food pairing was not investigated. It would appear that the pairing of foods is more complex and complicated than simply pairing foods that share common key compounds. Positive and negative synergistic and/or antagonistic volatile-volatile and volatile-matrix (volatile-lipid, volatile-protein and volatile-carbohydrate, etc.) interactions are important and must be taken into account. Additionally, positive and negative synergistic and/or antagonistic volatile-volatile interactions (odour intensity and flavour balance) are important aspects to consider when discussing food pairings.

An alternative hypothesis to the volatile sharing food pairing hypothesis would be that creating the right balance between odour active volatiles in foods is required to produce a volatile mixture that is perceived as pleasant or harmonious. It seems that the volatiles contributed by the banana may have provided bacon and basmati rice with the flavour and odour notes that they were lacking, and vice versa, producing a complete complementary flavour profile. In addition to this, the influence of non-volatile compounds contributing to the taste of the food parings (salty, sour, sweet, bitter and umami) must also be considered as an influential factor on hedonic evaluation. In particular, many of the assessors from the consumer panel noted that the "sweetness of the banana with the saltiness of the bacon worked well together" for $\mathrm{B}+\mathrm{BN}$. The
suitability of ingredients for inclusion in recipes or food pairings depends on a multitude of ingredient characteristics other than their flavour profile. Flavour is not necessarily the primary role of ingredients, recipes also rely on ingredients to provide final textures and overall structure of a given dish (Ahn et al., 2011). In addition, as mentioned by the participants of the focus groups, the flavour of a dish owes as much to the mode of preparation (cooking method) as to the choice of particular ingredients (This, 2005a; McGee, 2004).

### 5.7 Conclusions

The novel food pairings selected had complex flavour interactions which appeared to have influenced descriptive sensory evaluation and hedonic evaluation. Overall, two of the three novel food pairings ( $B+R$ and $B+B N$ ) were accepted by the consumer panel. It has been suggested in this chapter that possible complex positive and negative synergistic and antagonistic volatile interactions in the foods influenced the hedonic ratings of these food pairings. Based on these findings, it would be difficult to hypothesise that the success or failure of the food pairings in this chapter from a sensory perspective was due to the sharing or lack of sharing common volatiles alone. It appeared that creating certain flavour balances in volatile profiles of food pairings is important for a positive hedonic response. In addition, a binding phenomenon in the food matrices was more than likely an influencing factor on the rate of volatile release and the perceived aromas of the volatiles. Volatile concentration and odour thresholds are an important aspect to consider when discussing food pairings. Such knowledge may be exploited in the development of novel food products to produce interesting flavour combinations with a better understanding of consumer acceptance and rejection.

## CHAPTER 6

### 6.1 General introduction

New product development is a major competitive parameter for producers competing in mature and developed markets (Chen et al., 2010). However, food innovations can often be rejected by consumers as a result of a phobia towards novel foods (Barrena \& Sánchez, 2013). Humans principally learn to consume and prefer certain foods, and avoid and dislike others, with traditions, biological, psychological, and cultural factors being the main influences (Rozin, 2001). Few food preferences or aversions are innate; the majority of our food choices are learnt via differing degrees of exposure to foods (Nicklaus et al., 2004). Similar to food preferences, the development and maintenance of food acceptances are controlled by affective, personal, cultural and situational factors (de Klepper, 2011; Martins \& Pliner, 2005). Mere exposure, pairing of foods with positive or negative consequences and a variety of social influences cultivate a like or dislike for foods (Rozin, 2001). For these very reasons, food companies testing novel food products should identify the psychographic profiles of their consumer base to help explain potential variability in consumer response (Henriques et al., 2009). Consumer insight into the new product development process is important throughout the process itself (Grunert et al., 2011).

The main objectives of this chapter were to develop an ice cream recipe based on the results from chapters 3,4 and 5 , optimise the flavour of two novel flavoured ice creams, assess the consumer hedonic response to the ice cream products and explore consumer choice and preconceptions. Finally, the acceptability and suitability of these products for a market was investigated. The application of such knowledge for culinary industry practitioners and the food industry can be extremely beneficial.

### 6.2 Optimisation of ingredient concentration ratios

### 6.2.1 Banana and basmati rice ice cream concentration ratio

Numerous methods of basmati rice flavour transfer to the ice cream were tested using a preliminary sensory panel $(n=6)$ to determine the most appropriate method (see appendix D). It was found that boiling the basmati rice in the milk/cream mixture and immediately straining produced the most desirable results, and this was subsequently chosen as the appropriate method. The mean values for the hedonic scores of the five banana and rice ice creams $(B+R)$ formulations are presented in Table 6.1. A mean hedonic score of five was considered to be a limit of acceptance, all samples scored above this limit (>5).

Table 6.1 Mean hedonic scores with standard deviation and assessor comments (positive and negative) represented as a percentage (\%) of total number of subjects ( $\mathrm{n}=$ 34) for the banana and basmati rice ice cream formulations.

|  |  |  |  |  | Banana <br> comments \% |  | Basmati Rice <br> comments \% |  |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Sample | Banana <br> $(\mathbf{g} / \mathbf{k g})$ | Basmati <br> rice (g/ kg) | Hedonic <br> score | SD |  |  |  |  |
| Positive | Negative | Positive | Negative |  |  |  |  |  |

[^2]SD: standard deviation.

No significant differences ( $p>0.05$ ) were found between samples $B+R 3, B+R 4$ and $\mathrm{B}+\mathrm{R} 5$, which received mean hedonic scores of $7.1 \pm 1.4,7.0 \pm 1.2$ and $6.5 \pm 1.2$ respectively. As shown in Table 6.1, the lower levels of banana ( $150 \mathrm{~g} / \mathrm{kg}$ ) in B+R4 and B+R5 created a banana flavour that was described as "too mild", and for this reason was perceived negatively by the panellists ( $68 \%$ and $76 \%$ respectively). In contrast, the basmati rice flavour of $\mathrm{B}+\mathrm{R} 4(300 \mathrm{~g} / \mathrm{kg})$ was described as "too intense", and was perceived negatively (74\%). Thus, the characteristic banana fruitiness and sweetness was lacking in this formulation, which appeared to allow for the basmati rice flavour to overpower and dominate the flavour profile. In contrast to this, the low levels of both banana ( $150 \mathrm{~g} / \mathrm{kg}$ ) and basmati rice in $\mathrm{B}+\mathrm{R} 5(225 \mathrm{~g} / \mathrm{kg})$ appeared to produce an overall flavour that was perceived as 'too mild' and received negative feedback (91 \%). Conversely, B+R3 was described as having banana ( $225 \mathrm{~g} / \mathrm{kg}$ ) and basmati rice flavours $(300 \mathrm{~g} / \mathrm{kg})$ that were both "too intense" ( $82 \%$ and $85 \%$ respectively). Hence, a balance between banana and basmati rice flavour was not reached in these three formulations.

The higher scoring samples $B+R 1$ and $B+R 2(7.3 \pm 1.0$ and $7.7 \pm 1.0$ respectively) were not found to be significantly different ( $p>0.05$ ). Nevertheless, the qualitative data seemed to indicate that the flavour of $B+R 1$ was unbalanced. The majority of the assessors ( $68 \%$ ) noted that the flavour from the banana ( $225 \mathrm{~g} / \mathrm{kg}$ ) was "too intense". In contrast, the basmati rice flavour ( $150 \mathrm{~g} / \mathrm{kg}$ ) was described as "mild" $(85 \%)$. In addition to this, no significant differences were found between B+R1 and the ice cream formulations $\mathrm{B}+\mathrm{R} 3$ and $\mathrm{B}+\mathrm{R} 4$. While, $\mathrm{B}+\mathrm{R} 2$ was significantly different to all formulations except for $\mathrm{B}+\mathrm{R} 1$. What's more, the qualitative results indicate that sample $B+R 2$ had an apparent balanced combination of banana and basmati rice (both at 225 $\mathrm{g} / \mathrm{kg}$ ). The majority of panellists' comments were positive (74\%), mainly stating that it had a "good balance" of flavours.

Although the statistical analysis of the quantitative data could not yield a single formulation that was the significantly liked, it did offer an insight into directions to pursue (high banana and medium basmati rice levels) or avoid (lower banana and high basmati rice levels) in terms of a preferred ice cream formulation. Moreover, it can be concluded from the qualitative results that the banana content was the deciding factor in terms of perceived flavour balance with lower levels producing a flavour profile lacking in characteristic banana notes which, allowed the basmati rice flavour to dominate.

### 6.2.2 Banana and bacon ice cream concentration ratio

Several methods of bacon flavour transfer to the ice cream were tested using a preliminary sensory panel $(\mathrm{n}=6)$ to determine the most appropriate method (see appendix D ). The results showed that the method of soaking the cooked bacon in the milk/cream mixture for 24 hours produced the most desirable results, and was subsequently chosen as the appropriate method. The mean hedonic scores for the five different formulations of banana and bacon ice creams $(B+B N)$ are listed in Table 6.2.

No significances were found between samples $\mathrm{B}+\mathrm{BN} 3$ and $\mathrm{B}+\mathrm{BN} 4$. Both samples scored significantly ( $\mathrm{p} \leq 0.05$ ) lower hedonic scores ( $5.2 \pm 2.3$ and $4.8 \pm 2.1$ respectively) in comparison to the other formulations, with $\mathrm{B}+\mathrm{BN} 4$ scoring below the limit of acceptability (<5). The majority of the panellists provided negative comments ( $88 \%$ and $85 \%$ respectively) regarding the bacon flavour ( $300 \mathrm{~g} / \mathrm{kg}$ for both formulations), describing it as "too strong or intense" with a "salty" taste. Similarly, the banana flavour of these formulations ( $225 \mathrm{~g} / \mathrm{kg}$ for $\mathrm{B}+\mathrm{BN} 3$ and $150 \mathrm{~g} / \mathrm{kg}$ for $\mathrm{B}+\mathrm{BN} 4$ ) received negative feedback ( $56 \%$ and $88 \%$ respectively), and was described as "too mild".

Table 6.2 Hedonic scores with standard deviation and assessor comments (positive and negative) represented as a percentage (\%) of total number of subjects ( $\mathrm{n}=34$ ) for the banana and bacon ice cream formulations $(\mathrm{B}+\mathrm{BN})$.

| Samples | Banana <br> $(\mathbf{g} / \mathbf{k g})$ | Bacon <br> $(\mathrm{g} / \mathbf{k g})$ | Hedonic $\pm$ <br> score |  | Banana <br> comments \% <br> Positive |  | Bacon <br> comments \% |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Positive | Negative |  |  |  |  |  |  |  |

$\mathrm{B}+\mathrm{BN}(1-5)$ : banana and bacon ice cream formulations.
Samples with the different letters ( $\mathrm{a}-\mathrm{b}$ ) are significantly different ( $\mathrm{p} \leq 0.05$ ).

* optimum banana and bacon ice cream formulation.

SD: standard deviation.

No significant differences ( $\mathrm{p}>0.05$ ) were found between the ice cream formulations of $\mathrm{B}+\mathrm{BN} 1, \mathrm{~B}+\mathrm{BN} 2$ and $\mathrm{B}+\mathrm{BN} 5$, all scoring above the acceptability limit (> 5) $(6.6 \pm 1.7,6.5 \pm 1.5$ and $6.5 \pm 1.5$ respectively). It appeared that the flavour of $\mathrm{B}+\mathrm{BN} 1$ was perceived as unbalanced. Although the feedback for banana flavour (225 $\mathrm{g} / \mathrm{kg}$ ) was mainly positive ( $59 \%$ ), being described as "mild but nice", the bacon flavour received mainly negative feedback ( $94 \%$ ), and was noted as being "too mild". Similarly, B+BN5 received mainly negative feedback ( $68 \%$ for banana and $88 \%$ for bacon), the overall flavour ( $225 \mathrm{~g} / \mathrm{kg}$ for bacon and $150 \mathrm{~g} / \mathrm{kg}$ for banana) appeared to be "too mild".

Conversely, the feedback regarding banana and bacon flavour for $\mathrm{B}+\mathrm{BN} 2$ was mainly positive (65 \% and $71 \%$ respectively). This particular ice cream formulation consisted of equal levels of banana and bacon ( $225 \mathrm{~g} / \mathrm{kg}$ ) and many stated that the flavour was "balanced" with the bacon flavour "contributing pleasantly". The results seem to imply that the contribution of the bacon to the flavour profile of the ice cream
was the deciding factor in terms of perceived flavour profile. It would appear that at higher concentrations ( $300 \mathrm{~g} / \mathrm{kg}$ ), an unpleasant salty taste accompanied the distinctive bacon flavour, which appeared to prevail regardless of the banana levels.

### 6.2.3 Salt and pH analysis of optimised ice cream formulations

The salt $(\mathrm{NaCl})$ content of the optimised ice cream formulations from section 6.2.1 ( $\mathrm{B}+\mathrm{R} 2$ ) and $6.2 .2(\mathrm{~B}+\mathrm{BN} 2)$ was measured as results from chapter 4 showed that salt content was a significant influencing variable on the emission of volatiles from emulsion products. In addition to this, the pH levels of the ice cream formulations were measured to ensure that they were in the optimum ranges (chapter 4). The mean salt concentrations and pH levels for banana and bacon, banana and rice, control 1 (banana $225 \mathrm{~g} / \mathrm{kg}$ ) and control 2 (plain) ice creams and their significant levels are displayed in Table 6.3.

Table 6.3 Mean Salt concentration (g/ 100 g ) and pH levels of ice creams and control formulations.

| Ice cream formulations | Salt conc. $(\mathbf{g} / \mathbf{1 0 0} \mathbf{g})$ | $\mathbf{S D}$ | $\mathbf{p H}$ | SD |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Banana and bacon | $3.34^{\mathrm{a}}$ | $\pm 0.08$ | $6.07^{\mathrm{a}}$ | $\pm 0.05$ |
| Banana and basmati rice | $0.04^{\mathrm{b}}$ | $\pm 0.01$ | $6.24^{\mathrm{b}}$ | $\pm 0.04$ |
| Control 1 | $0.05^{\mathrm{c}}$ | $\pm 0.02$ | $6.05^{\mathrm{a}}$ | $\pm 0.01$ |
| Control 2 | $0.06^{\mathrm{d}}$ | $\pm 0.02$ | $6.38^{\mathrm{c}}$ | $\pm 0.01$ |

Samples with different letters ( $\mathrm{a}-\mathrm{d}$ ) have significantly different means within each column ( $\mathrm{p} \leq 0.05$ ). Control 1: banana ice cream.
Control 2: plain ice cream.
SD: standard deviation.
Significant differences ( $\mathrm{p} \leq 0.05$ ) were found between the salt concentrations of the ice cream samples. Banana and bacon was found to have a significantly higher ( $\mathrm{p} \leq$ $0.05)$ salt level $(3.34 \pm 0.08 \mathrm{~g} / 100 \mathrm{~g})$ compared to all other samples as would be
expected due to the relatively high concentration in the bacon itself ( $3.4 \mathrm{~g} / 100 \mathrm{~g}$ ). In addition, numerous significant differences were found between the pH levels of the ice creams. Control 2 (plain ice cream) had a significantly higher $(\mathrm{p} \leq 0.05) \mathrm{pH}(6.38 \pm$ $0.01)$ compared to the other ice cream formulations. The addition of banana appeared to significantly lower ( $\mathrm{p} \leq 0.05$ ) the pH in control 1 (banana ice cream, $6.05 \pm 0.01$ ) compared to control 2 . No significant differences ( $\mathrm{p}>0.05$ ) were found for the pH of banana and bacon ice cream and control 1, implying that bacon had no influence on pH . Conversely, the banana and basmati rice ice cream had a significantly higher pH ( $\mathrm{p} \leq$ $0.05)(6.24 \pm 0.04)$ to control 1 and banana and bacon ice cream. This would suggest that the addition of basmati rice influenced the pH of this ice cream formulation by increasing the pH .

### 6.3 Consumer acceptance and market suitability

The mean hedonic scores and standard deviations for evaluation of banana and basmati rice ice cream and banana and bacon ice cream by a sensory panel representing the general retail market (panel A) and the panel representing the culinary educated market (panel B) are presented in Table 6.4. Both panels (panels A and B) provided comparable mean hedonic scores for the banana and rice ice cream $(B+R)$, with no significant differences $(p>0.05)$ found $(7.2 \pm 1.2$ and $7.0 \pm 1.2$ respectively). In addition to this, a relatively narrow range of the hedonic scales were used by both panels (panel A, 4.5-9 from lowest - highest and panel B, 4.2-9 from lowest to highest). The feedback for this ice cream was mainly positive ( $90.8 \%$ for both panels), with the many assessors stating that the ice cream had a "pleasant flavour" that was "unique and interesting". While, $83 \%$ of panellists from panel A and $82 \%$ from
panel B stated that if banana and basmati rice ice cream ( $B+R$ ) was available on the market that they would be willing to purchase it.

Table 6.4 Mean hedonic scores with standard deviation and assessor comments (positive and negative) represented as a percentage (\%) of total number of subjects ( $\mathrm{n}=$ 76) for each sensory panel (panel A and panel B) for banana and basmati rice ice cream and banana and bacon ice cream.

| Sample | Panel | Hedonic score | SD | Positive comments \% | Negative comments \% |
| :---: | :---: | :---: | ---: | :---: | :---: |
| B+R | Panel A | $7.2^{\mathrm{a}}$ | $\pm 1.2$ | $90.8 \%$ | $9.2 \%$ |
| B+R | Panel B | $7.0^{\mathrm{a}}$ | $\pm 1.2$ | $90.8 \%$ | $9.2 \%$ |
| B+BN | Panel A | $5.1^{\mathrm{b}}$ | $\pm 2.3$ | $27.6 \%$ | $72.4 \%$ |
| B+BN | Panel B | $6.8^{\mathrm{a}}$ | $\pm 1.2$ | $65.8 \%$ | $34.2 \%$ |

[^3]In contrast to this, banana and bacon ice cream ( $\mathrm{B}+\mathrm{BN}$ ) was liked significantly less $(\mathrm{p} \leq 0.05)$ by panel A than by panel B, $5.1 \pm 2.3$ and $6.8 \pm 1.2$ respectively (Table 6.3). In addition, a much wider range of the hedonic scale was used by panel A (0.3-9 from lowest to highest). It is clear from this usage of a wider range of the hedonic scale and the relatively large standard deviation, that there was greater variation in the perception of $\mathrm{B}+\mathrm{BN}$ amongst the assessors of the panel A . Moreover, the feedback for B+BN from panel A was mainly negative (72.4 \%), with many stating that they "didn't like the unusual taste" and found it too "salty for an ice cream". On the contrary, a narrower range of the hedonic scales was used by panel B (4.5-9 from lowest to highest), implying less variation amongst assessor's perception of B+BN. What's more, the feedback for $\mathrm{B}+\mathrm{BN}$ from panel B was mainly positive ( $65.8 \%$ ), with many of the
assessors noting that they "liked the sweet and salty combination" and they found the flavours to be "very interesting" and "unique". Only $46 \%$ of panellists from panel A expressed a willingness to purchase the $\mathrm{B}+\mathrm{BN}$ if it was available on the market, whereas, $79 \%$ of panellists from panel $B$ expressed a willingness to purchase $B+B N$.

From the results, two clear differences can be seen in the hedonic assessment of the ice creams: (1) a difference between the assessment of $\mathrm{B}+\mathrm{BN}$ by panel A and panel $B$ and (2) a difference between the assessment of $\mathrm{B}+\mathrm{R}$ and $\mathrm{B}+\mathrm{BN}$ by panel A . The difference between the assessment of $\mathrm{B}+\mathrm{BN}$ by panel A and panel B between may be explained by the type of assessors in panel A. This panel represented the general retail market, and consisted of subjects who more than likely had varying limited exposure to novel food pairings, which seems to have triggered neophobic behaviour (Henriques et al., 2009). In contrast, panel B consisted of subjects who due to their careers and/or educational background in a culinary discipline, would have more exposure to the concept of novel food pairings, reducing the likelihood of neophobic behaviour being triggered. It has been noted that mere exposure and increasing familiarity with unfamiliar foods may play an important role in the food preference development and reduction of neophobia (Reverdy et al., 2010). Birch \& Marlin (1982) and Pliner (1982) demonstrated that preference increased with exposure frequency to novel foods.

The differences in hedonic assessment of $B+R$ and $B+B N$ by panel $A$ could be explained by the main flavour being a food of animal origin (bacon) in B+BN. This may have caused a neophobic response of dislike, described by Martins et al., (1997) as a negative pole of the Ideational dimension. Similarly, Martins \& Pliner (2005) found that participants in sensory panels were more neophobic with respect to novel animal foods than novel non animal foods, with stronger disgust responses and less willingness to try animal products. Essentially, these foods are rejected because of what they are, where
they come from, or their social history e.g. who touched them or ate them (Martins \& Pliner, 2006). Furthermore, it is likely that this neophobic behaviour was a predisposed response, formed prior to consumption of the ice cream due to an anticipated dislike of the ice cream (Raudenbush \& Frank, 1999). Arvola et al. (1999) found that neophobic subjects rated expected and actual taste pleasantness lower to neophilics for unfamiliar cheeses. Also, it may be the case that the more sweet and savoury tasting B+R was simply more appealing to panel A than the sweet, salty and meaty flavour of B+BN. Neophilics are more willing to try products that may be stronger in flavour characteristics, whereas neophobics tend to select flavours with sweeter flavour profiles (King et al., 2008).

These results would suggest that if $\mathrm{B}+\mathrm{BN}$ was released on the general market that it may not compete successfully with market dominant ice cream brands. In contrast to this, $\mathrm{B}+\mathrm{BN}$ would appear to be a product that may perform better in an environment where novel flavour pairings is more common and expected as a unique selling point, such as a higher end restaurant or in culinary specialised outlets. The banana and basmati rice ice cream offered an alternative ice cream product to banana and bacon ice cream, where the main flavour wasn't animal based (bacon), thereby avoiding the possible neophobic reaction related to some products of animal origin. In addition to this, perception of a strong salt taste related to higher bacon levels (section 6.2.3) proved to be a factor which influenced hedonic rating of the products. Therefore, banana and basmati rice ice cream offered an alternative to a salty tasting product with salt levels that were low $(0.04 \pm 0.01 \mathrm{~g} / 100 \mathrm{~g})$. These factors may explain this products likelihood of being more viable in a competitive general market than banana and bacon ice cream. Additionally, customers of such establishments would be trusting in the chef's ability to successfully pair novel foods in a manner that was appetising and
appealing. It is worth noting that food manufacturers assess marketplace trends, and very often monitor what trends are currently popular in restaurants in order to develop concepts for new products (Moskowitz et al., 2009).

### 6.4 Conclusions

It was found that bacon flavour had a profound influence on the perception of a banana and bacon ice cream. On the other hand, banana flavour was found to be the influential factor in the perception of banana and basmati rice ice cream. The banana and basmati rice ice cream was accepted by a sensory panel representing the general retail market (panel A) and by a sensory panel representing a culinary educated market (panel B), suggesting it would perform well in both market places. In contrast, apparent neophobic behaviour developed during the assessment of banana and bacon ice cream by panel A which was more than likely a result the panellists lacking exposure to novel food pairings. For this reason, this product may be more successful in high end restaurants as opposed to in a commercial market. It is an interesting finding that there were significant differences between ice creams with ingredients of animal origin (bacon) and ice creams without. Another important finding is that not only was the hedonic evaluation of these food products influenced by the actual products being evaluated (banana and bacon or banana and basmati), but is also influenced by the subjects and their exposure to such novelty who made up the sensory panels.

The effect of salt concentration on the flavour release and sensory perception in the ice cream products was not evaluated in this study. Hence, there is potential for this to be investigated specifically in the banana and basmati rice ice cream product. This product would be a good candidate for such a study as it has naturally low levels of salt.

Comparison of a sample of little or no salt levels with samples of different levels of salt may have revealed results that further corroborated the results of chapter 4 where flavour perception was enhanced by increasing salt levels. In addition to this, it may have also revealed further mechanisms of phenomena that could have influenced sensory hedonic responses.

## Chapter 7

### 7.1 General discussion

Product development is a process of creativity and discovery that intends to accumulate knowledge, create something new from trial-and-error and learn from the errors made (Smith \& Eppinger, 1997). Each development activity is responsible for generating the required product or process knowledge based on the input information received from other activities. The information evolution of a development activity is considered as a gradual refinement of the product or process knowledge from its preliminary form (prototypes) to a final end product (Wang \& Lin, 2009). In this work, a molecular gastronomy approach to food product development using methods of investigation of the physical and chemical culinary mechanisms of phenomena was applied to gradual refine the product. This approach allowed for the gradual refinement of food products and the processing of knowledge obtained from experimental investigation of fundamental and important culinary phenomena which contribute to sensory experiences. Two novel ice cream products were developed through a holistic scientific approach to food product development, where phenomena that occurred on a molecular level right through to phenomena that occurred on a psychological level were investigated.

Each results and discussion chapter consisted of an investigation of different but related culinary mechanisms of phenomena, providing an improved scientific understanding and insight. Such mechanisms included physical stability (chapter 3), flavour release (chapter 4), flavour volatile interactions influencing sensory perception (chapter 5) and consumer perception and acceptance (chapter 6). The common objective of each chapter was to apply the knowledge obtained through the experimental work to improve the overall sensory experience of the developed novel food product. Thus, this study was in line with the aims and objective of molecular gastronomy, where
maximising the consumer's sensory experience is paramount (Barham et al., 2010; This \& Rutledge, 2009; van der Linden, 2008; This, 2005a; This 2005b).

The first stage of the development process, described in chapters 3 and 4, involved the exploration of some of the primary factors which influence the formation and stability of emulsion based food products, and subsequently the release of two volatiles from these dispersions. One of the most important factors determining the commercial viability of food products is the ability to resist changes in their physical properties after production. Optimising the physical stability of food products is crucial in order to manipulate textures and flavours, create visually appealing food products and increase shelf life (Piorkowski \& McClements, 2013). For these reasons, one of the major objectives of food product developers is to establish the factors which determine the stability of a food product, as well as to elucidate general principles which can be used to predict the behaviour of new products or processes (McClements, 2005).

Computer modelling techniques have been used to investigate the relationship among colloidal interactions (Gharibzahedi et al., 2012; Lorenzo et al., 2008; Mirhosseini et al., 2008a; Mirhosseini et al., 2008b; Pey et al., 2006; Buffo et al., 2002; Wijmans, et al., 1999). The interactions between emulsion ingredients (sunflower oil, lecithin and xanthan gum) were investigated using RSM (chapter 3). Optimum conditions for maximum emulsion storage stability and minimum mean droplet diameter were established through this regression modelling. The creation of a food model system that fully represents an extremely complex food product like ice cream is a difficult challenge. Thus, the creation of a major component (an emulsion) of an ice cream product allowed for the testing of culinary mechanisms of phenomena which may occur within this product.

Colloidal design offers much potential for producing tastier more flavoursome food. Improvements in understanding emulsion stabilisation and destabilisation is a means of targeting delivery of flavours (Douaire \& Norton, 2013). Hence, once the formation and stability of emulsion model systems was optimised, the effects of physicochemical parameters ( pH and salt concentration) on the volatile release of commonly used banana flavour compounds (isoamyl acetate and furfuryl acetate) from the emulsion were investigated in chapter 4 . Flavour is one of the most important factors determining the perceived quality of foods, and consumers expect that each type of food product will have its own particular characteristic flavour profile (Gilbert \& Firestein, 2002). McClements (2005) notes that flavour partitioning and mass transport in emulsions can depend on the physicochemical properties of the component phases (e.g., polarity, rheology, and physical state). Therefore, establishing the optimum physicochemical conditions for maximum transfer of volatile compounds to the olfactory receptors in the human nose is of utmost importance (van Ruth \& Roozen, 2000).

Application of static headspace and dynamic methods has given insight into the partitioning of flavours between the liquid and the gaseous phase (Piggott \& Schaschke, 2001). Apart from experimental data, static and dynamic flavour release from food matrices were successfully modelled (de Roos, 2000). Empirical models can lead to predictions of volatile partitioning and dynamic flavour release (Taylor \& Linforth, 2001; Katritzky et al., 1998). In this work, RSM was employed to model volatile emission from the food emulsion model system, to examine the interactions between volatile compounds and physicochemical variables and to establish the optimal conditions for maximum flavour release. As with the results from chapter 3, they can only be partially incorporated into the finally product as the model did not completely
represent an ice cream. The pH of the ice creams ( 6.07 for banana and rice ice cream and 6.34 for banana and bacon ice cream) corresponded with the optimal pH (6.02) levels established in chapter 4 . The banana and bacon ice cream had relatively high levels of salt ( $3.34 \% \mathrm{w} / \mathrm{v} \pm 0.08$ ) which allowed for a significant increase ( $\mathrm{p} \leq 0.05$ ) in volatile emission.

The second stage of the development process, described in chapters 5 and 6 , involved building on and integrating the results from chapters 3 and 4 to produce novel flavoured ice cream products which ultimately were acceptable to the consumer. A food product developer must ensure that the flavour profile of a product is desirable and that it conforms to consumer expectations for that kind of product (McClements, 2005). Results from the consumer evaluation of the novel pairing showed that banana and basmati rice and banana and bacon were acceptable food pairings while banana and extra virgin olive oil was not (chapter 5). The work carried out in this chapter involved looking at the culinary phenomena of food pairing and is of great importance to food product development which is ultimately concerned with consumer hedonic response to food. Furthermore, very little work has been published in this area, further reiterating the importance of the study and the importance of further investigation of the subject area.

In product development, instrumental measurements (GC/MS) are frequently coupled with sensory analysis techniques (sensory profiling) to try and determine the exact volatile responsible for some flavour sensations (Chambers \& Koppel, 2013). In this study, the correlation of descriptive analysis data with instrumental volatile profiling data provided a better understanding of the hedonic results for the food pairings and revealed the complexity of the food component interactions influencing sensory perception. The design of foods with desirable flavour profiles depends on an
understanding of the relationship between the types and concentrations of flavouring substances present and the final flavour perceived by consumers (Pothakamury \& Barbosa-Canovas, 1995).

The results thus far led to the production of the final ice cream products, banana and basmati rice and banana and bacon ice creams (chapter 6). Flavour is more than just a series of complex chemical interactions in foods, but is also an interaction of the food and the consumer. For the successful introduction of innovations in traditional food products like ice creams, it is also important to have a good understanding of consumers' perceptions, expectations and attitudes towards innovating traditional food products (Linnemann et al., 2006). This ensures that the product is in line with consumer preferences, while all the time being innovative and cutting edge.

Furthermore, the constant generation of new knowledge through scientific investigation of chemical and physical interactions in food products in tandem with consumer integration throughout the development process of the ice creams was important for optimisation and streamlining of the process. This can be extremely vital for innovation as it accelerates the progress of development projects which can lead to premium pricing and higher sales volume for a new product (Langerak \& Hultink, 2005). Increasing the speed of new product development allows for continuous reduction in the product life cycle time and increase in competition from technological advancements and globalisation (Chen et al., 2010). Food companies can achieve several important benefits from increasing the speed of new product development; first, increased profitability, margins, and market share (Brown \& Eisenhardt, 1995). Second, companies with faster new product development have a greater chance to establish industry standards and may lock up distribution channels (Dumaine, 1989). Third, a company with the capability of developing products rapidly can quickly respond to
market demands, improving the timeliness of its product entry and customer satisfaction (Chen et al., 2010).

Food product developers wish to know the type and amount of flavouring components that must be incorporated into a food during the manufacturing process in order to produce a desirable flavour profile in the final food product. On the other hand, a food manufacturer may want to know how to avoid the production of an off-flavour within a food during manufacture, storage, or usage (McClements, 2005). For these reasons, consumer testing and market research were required on the final ice cream products to establish preferred ice cream formulations and to assess their overall acceptability (chapter 6).

Results showed that not only is the hedonic evaluation of food products influenced by the actual products being evaluated, but also by the subjects evaluating them and by their level of exposure to such novelties. Predispostional neophobic behaviours can be factors which impede the commercial growth of a product in a general market and must be overcome. This appeared to be the case for banana and bacon ice cream. However, banana and basmati rice ice cream offered an alternative ice cream product, one with more potential viability in a competitive general market. This further reiterates the importance of customer integration in the development process. The implications of these results to the development of food products can be huge. Food product developers must not only assess if a product is acceptable or not, but also investigate the reasons why and address them if the product is considered unacceptable or is rejected. Moreover, some products may only be initially suitable for specialised markets, and exposure of the product to broader market may also bring success in a more general market at a later stage.

Through using a comprehensive molecular gastronomy approach, the new product development process in this study was optimised in order to produce high quality products which satisfy the rapidly evolving and increasing demands of consumers.

### 7.2 Key outcomes

- Emulsion ingredients had varying degrees of positive and negative influences on emulsion stability and mean droplet diameter over 14 days of storage.
$>$ High levels of xanthan gum ( $0.28 \% \mathrm{w} / \mathrm{v}$ ), low levels of lecithin ( $1.2 \%$ $\mathrm{w} / \mathrm{v}$ ) and high levels of sunflower oil ( $19.02 \% \mathrm{w} / \mathrm{v}$ ) resulted in optimum emulsion stability ratio ( $100 \%$ ) and mean droplet diameter ( $4.35 \mu \mathrm{~m}$ ).
- Isoamyl acetate, furfural acetate and salt concentrations had significant influences on the headspace emission of isoamyl acetate from the emulsion.
$>$ Increasing salt concentration subsequently resulted in an increased release of isoamyl acetate into the headspace of the emulsions.
> Increasing isoamyl acetate concentration subsequently resulted in a linear increase of release of isoamyl acetate into the headspace of the emulsions.
> Increasing furfuryl acetate concentration subsequently resulted in a reduction of isoamyl acetate headspace release.
- The novel food pairings selected had complex volatile interactions which appeared to have influenced descriptive sensory evaluation and hedonic evaluation.
> Banana and basmati rice and banana and bacon food pairings were accepted by the panel of consumers, while banana and extra virgin olive oil was not accepted.
- Complex synergistic volatile interactions in the foods appeared to have influenced the hedonic ratings of these food pairings.
$>$ A possible volatile-volatile interaction regarding odour intensity may have occurred.
> A possible volatile-volatile interaction may have involved the odour active compounds of the constituents of the food pairings
$>$ These possible interactions proved to be positive for the pairings of banana and basmati rice and banana and bacon, resulting in harmonious and pleasant organic volatile mixture and a positive hedonic response.
$>$ These possible interactions proved to be negative for the pairing of banana and extra virgin olive oil, resulting in unpleasant organic volatile mixture and a negative hedonic response.
- The ice creams were perceived differently by the panels representing different markets.
$>$ Hedonic evaluation was greatly influenced by the subjects who made up the sensory panels.
$>$ Predispositional neophobic behaviour appeared to be present in the perception of banana and bacon ice creams in the panel representing the general market.


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## Appendices

## Appendix A:

## Sensory analysis documentation

## Sensory analysis consent form



Please note:

- For persons under 18 years of age the consent of the parents or guardians must be obtained or an explanation given to the Research Ethics Committee and the assent of the child/young person should be obtained to the degree possible dependent on the age of the child/young person. Please complete the Consent Form (section 4) for Research Involving 'Less Powerful' Subjects or Those Under 18 Yrs.
- In some studies, witnessed consent may be appropriate.
- The researcher concerned must sign the consent form after having explained the project to the subject and after having answered his/her questions about the project.


## Sensory analysis information sheet

Project aim: To carry out sensory analysis on novel flavour combinations in order to determine if the products are acceptable to the consumer.

Project information: Our research is being conducted as part of a PhD project on development of innovative novel food products using the theories and techniques of molecular gastronomy. You will be assessing the acceptability of three samples of foods in combination with banana in terms of taste in order to determine if these products are acceptable to consumers. If you have any allergies with the following foods: bacon, olive oil, basmati rice and banana please inform the researchers. No extra ingredients have been added.

If there is any reasonable doubt or uncertainty about the safety of the material you will not be asked to taste it. Participation in the study is strictly voluntary and participants can withdraw at any time, without giving a reason. If you are a diabetic, or have a cold or sinusitis please do not participate in this taste test. If you have any illness which you feel may compromise your health please do not participate in this test.

The results will be presented in the PhD thesis and are anonymous.

If you have any further questions you can contact Mark Traynor at mark.traynor@dit.ie, (01) 402 4373. This project has been approved by the DIT Research Ethics Committee who can be contacted at researchethicscommittee @ dit.ie at any time.

## Appendix B:

## Sensory analysis evaluation sheets

## Sensory evaluation of orthonasal acceptability of food pairings test

## Instructions:

Read the instructions of this sensory analysis test very carefully.

You will be presented with eight randomly coded samples, one at a time. You are requested not to open the container. Smell the samples carefully through the holes in the lids, take three short sniffs. Complete the assessment of this odour before proceeding to the next sample.

Please rate the samples aroma acceptability by placing a tick opposite the appropriate term.

| Sample | Extremely <br> Like | Like <br> Very <br> Much | Moderately <br> Like | Like <br> Slightly | Neither <br> Like or <br> Dislike | Dislike <br> Slightly | Moderately <br> Dislike | Dislike <br> Very <br> Much | Extremely <br> Dislike |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| 170 |  |  |  |  |  |  |  |  |  |
| 463 |  |  |  |  |  |  |  |  |  |
| 549 |  |  |  |  |  |  |  |  |  |
| 774 |  |  |  |  |  |  |  |  |  |
| 289 |  |  |  |  |  |  |  |  |  |
| 320 |  |  |  |  |  |  |  |  |  |
| 953 |  |  |  |  |  |  |  |  |  |
| 641 |  |  |  |  |  |  |  |  |  |

## Sensory evaluation of taste acceptability of food pairings test

Instructions:

Read the instructions of this sensory analysis test very carefully.

You will be presented with three coded samples. The samples will be presented to you separately and one at a time, please only rate the acceptability of the sample's flavour by placing a tick opposite the appropriate term. After rating the acceptability, you will be asked to answer a question on each sample. Once you have assessed a sample, this sample will be removed and the next sample for evaluation will be presented. Please cleanse your palate with the crackers and water provided in between samples.

Please do not communicate with any other participants while taking part in the sensory analysis. If you have any questions during the sensory analysis please do not hesitate to ask one of the researchers.

Thank you.

Sample 321

Please rate the sample acceptability by placing a tick opposite the appropriate term

| Extremely | Dislike Very | Moderately | Dislike | Neither Like | like | Moderately | like Very | Extremely |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| Dislike | Much | dislike | Slightly | or Dislike | Slightly | like | Much | like |

Do you think the pairing of these two foods works?

Any further comments regarding this food pairing?

## Preference ranking test for food pairings

Instructions:

You have been presented with a set of 3 randomly coded samples. Please taste the samples and use the water provided to cleanse your palate before tasting each sample:

## 321471928

Place the code numbers in the appropriate position below. One code only per line - no ties are allowed).

| Preference | Codes |
| :--- | :--- |
| Most preferred |  |
| Second most preferred |  |
| Least preferred |  |

## Appendix C:

## Free choice profiling evaluation sheets and descriptor lexicons

Orthonasal and retronasal descriptive lexicons for food pairings

| BANANA AND BACON |  |
| :---: | :---: |
| AROMA | FLAVOUR |
| Sweet | Sweet |
| Sugary | Sweet, caramel |
| Caramelised | Burnt sugar |
| sweet caramel | Maple |
| Maple | Savoury |
| Salty | Umami |
| roasted meat | Salty |
| Meat | roasted meat |
| Fried meat | Meaty |
| Umami | fried meat |
| Savoury | Fatty |
| Greasy | Oily |
| Fatty | Greasy |
| Greasy | Fruity |
| Oily | Floral |
| Buttery | Lemon |
| Smoky | Sweet, fruity |
| Popcorn | Nutty |
| Bready | Bready |
| Earthy | Popcorn |
| Floral | Cereal |
| Green floral | Smoky |
| Pear | Woody |
|  | Pear |

Orthonasal evaluation of food pairings by Free Choice Profiling

PANELLIST NAME: $\qquad$

SAMPLE CODE: $\qquad$


Descriptive term: $\qquad$


Descriptive term: $\qquad$


## Appendix D:

# Determination of flavour transfer 

## method for ice creams study

## D. 1 Introduction

This appendix comprises of the preliminary study into the determination of the most appropriate method of flavour transfer for basmati rice and bacon to the respective ice creams. Numerous methods of flavour transfer were tested for both ice creams. The flavours were incorporated into a mixture of full fat milk (3 \% v/v fat) and cream ( $38 \%$ $\mathrm{v} / \mathrm{v}$ fat) at a ratio of 52.8:47.2 \% (v/v). As the milk/cream mixture was the main ingredient in the ice creams it was evaluated as opposed to actually ice creams. The selected basmati rice and bacon contents were $225 \mathrm{~kg} / \mathrm{L}$ for both. A sensory panel ( $\mathrm{n}=$ 6) was used to assess the milk/cream mixtures. The factors considered were: (1) the intensity of the basmati or bacon taste, (2) off tastes or undesirable textures, and (3) the physical state of the mixtures.

## D. 2 Materials and methods

## D.2.1 Materials

Raw materials are as described in section 2.5.1.

## D.2.2 Methods

D.2.2.1 Banana and basmati rice ice cream flavour transfer

Six methods of flavour transfer methods for basmati rice to the banana and basmati rice ice cream were tested in total. The basmati rice to milk/cream mixture ratio was 225 g : 1 L for these tests. The flavour transfer methods were as follows:

- Method R1: Boil and strain: raw basmati rice was cooked for 15 minutes in the milk/cream mixture. The rice was then strained immediately after boiling and discarded, and the cooking liquid reserved.
- Method R2: Boil and soak: raw basmati rice was cooked for 15 minutes in the milk/cream mixture. The rice was soaked in the milk/cream mixture for 24 hours at approximately $4{ }^{\circ} \mathrm{C}$. The rice was then strained and discarded, and the cooking liquid reserved.
- Method R3: Grind, boil and strain: raw basmati rice was ground to a course powder in a coffee grind (model). The basmati rice coarse powder was then boiled in the milk/cream mixture for 15 minutes. The rice was strained immediately after boiling and discarded, and the cooking liquid reserved.
- Method R4: Magnetic hotplate stirrer: raw basmati rice was heated in the milk/cream mixture in a glass beaker on a magnetic stirrer hotplate (Stuart CB162, Bibby-scientific, Staffordshire, UK) at 100 rpm at $60^{\circ} \mathrm{C}$ for 15 minutes. The rice was then strained immediately after heating and discarded, and the cooking liquid reserved
- Method R5: Incubation agitation: raw basmati rice was heated in the milk/cream mixture in a glass beaker in an incubator (Innova 42, New Brunswick Scientific, CT, USA) at 100 rpm at $60{ }^{\circ} \mathrm{C}$ for 15 minutes. The rice was then strained immediately after heating and discarded, and the cooking liquid reserved.
- Method R6: Grind and hot plate stirrer: raw basmati rice was heated in the milk/cream mixture in a glass beaker on a magnetic stirrer hotplate (Stuart CB162, Bibby-scientific, Staffordshire, UK) at 100 rpm at $60^{\circ} \mathrm{C}$. The rice was then strained immediately after heating and discarded, and the cooking liquid reserved.
D.2.2.2 Banana and bacon ice cream flavour transfer

Bacon was cooked as described in section 2.4.2.3. Four methods of flavour transfer for bacon to the banana and bacon ice cream were tested in total. The bacon to milk/cream mixture ratio was 225 g : 1 L for these tests. The flavour transfer methods were as follows:

- Method B1: Soak and strain: cooked bacon was soaked in the milk/cream mixture for 24 hours at $4^{\circ} \mathrm{C}$. The bacon was strained and discarded, and the soaking liquid reserved.
- Method B2: Boil and strain: cooked bacon rice was cooked for 15 minutes the milk/cream mixture. The bacon was then strained immediately after boiling and discarded, and the cooking liquid reserved.
- Method B3: Boil and soak: cooked bacon was cooked for 15 minutes in the $\mathrm{milk} /$ cream mixture. The bacon was soaked in the milk/cream mixture for 24 hours at approximately $4{ }^{\circ} \mathrm{C}$. The bacon was then strained and discarded, and the cooking liquid reserved.
- Method B4: Salt removal and soak: cooked 225 g of bacon was soaked in 2 litres of distilled, deionized water (DDW) from a Milli-Q water purification system (Millipore, Bedford, MA, USA) for 24 hours. The bacon was then strained and soaked the milk/cream mixture for a further 24 hours at approximately $4^{\circ} \mathrm{C}$. The bacon was then strained and discarded, and the cooking liquid reserved.
D.3.2.3 Sensory evaluation of the flavoured milk and cream mixtures

Sensory evaluation of the basmati rice flavoured the milk/cream mixtures and of the bacon flavour the milk/cream mixture were assessed by a sensory panel $(\mathrm{n}=6)$. Sensory evaluation was carried out in a sensory laboratory under guidelines and conditions according to ISO 8589:2010 (ISO, 2010). The panellists worked in a single booth under defined conditions of $22{ }^{\circ} \mathrm{C}$ and white light. Panellists were presented with 10 ml samples a monadic sequential order to the panel. Panellists were instructed to evaluate taste acceptability of the paired samples on a nine point hedonic scale, where 9 $=$ "like extremely", $5=$ "neither like nor dislike" and $1=$ "dislike extremely" (see appendix B). Additionally, panellists were encouraged to write comments regarding their opinion of the milk and cream mixtures. Sensory evaluation of the basmati rice and bacon flavour transfer methods took place on separate days.

## D.3.2.4 Physical state assessment

The physical state of the flavoured the milk/cream mixtures was assessed by visual inspection. The change of the thickness (viscosity) of the samples was assessed by visual and touch (mouthfeel) inspection. The overall end product yield from 500 ml of the milk and cream mixture was measured using a graduated cylinder.

## D. 3 Results and discussion

## D.3.1 Basmati rice flavour results

A summary of the results from the assessment of basmati rice flavours transfer methods from rice to the milk/cream mixture are presented in Table D.1. Due to the hydrophilicity of the rice, this method of boiling and soaking the rice (method R2) produced no end product yield to be assessed, hence this method produced the least desirable results. Method R4 and method R5 produced slightly viscous end products with the same approximate yield ( 300 ml ) with mild rice flavours. On the other hand, the method of grinding the rice to a coarse powder (method R3 and method R6) produced extremely small yields of end product ( 50 ml ) which were very viscous and partially solid with mild rice flavours. From the results it was decided that method R1 (boil and strain) was the most appropriate method for transferring the rice flavour to the milk/cream mixture. It produced the highest mean acceptability scoring ( $8 \pm 1$ ), the highest overall yield ( 350 ml ), while its thickness only increased slightly. Additionally, the assessors stated that it had a strong characteristic rice flavour in comparison to the other samples.

Table D. 1 Summary of the results for the assessment of basmati rice flavour transfer methods from rice to the milk/cream mixture.

| Method | Mean hedonic <br> score | Approx. yield (ml) <br> from $\mathbf{5 0 0} \mathbf{~ m l}$ | Thickness | Summary of <br> comments |
| :---: | :---: | :---: | :---: | :---: |
| Method R1 | $8 \pm 1$ | 350 ml | Slightly viscous | Strong rice flavour |
| Method R2 | NA | 0 | Thick and solid | NA |
| Method R3 | $5 \pm 1$ | 50 ml | Thick and semi-solid | Mild rice flavour |
| Method R4 | $4 \pm 1$ | 300 ml | Slightly viscous | Mild rice flavour |
| Method R5 | $3 \pm 1$ | 300 ml | Slightly viscous | Mild rice flavour |
| Method R | $6 \pm 1$ | 50 ml | Thick and semi-solid | Mild rice flavour |

## D.3.2 Bacon flavour results

A summary of the results from the assessment of bacon flavours transfer methods from bacon to the milk/cream mixture are presented in Table D.2. From the results it can be seen that boiling the bacon in the milk/cream mixture (method B2 and method B3) produced the strongest bacon flavours, however it also produced a strong and unpleasant salt taste. Due to this, these methods of flavour transfer produced the lowest mean hedonic scorings $(2 \pm 1)$ and the least desirable overall results. In contrast to these methods, method B4 which involved removal of the salt through soaking in water produced extremely mild bacon flavour and salt taste, and so this method was not appropriate. It can be seen that the method of soaking the bacon for 24 hours in the milk and cream mixture and straining it (method B1) produced a mild bacon and salt taste, with the highest mean hedonic scoring ( $7 \pm 1$ ), and was therefore the most appropriate method of transferring the bacon flavour to the milk/cream mixture.

Table D. 2 Summary of the results for the assessment of bacon flavour transfer methods from bacon to the milk/cream mixture.

| Method | Mean hedonic <br> score | Approx. yield <br> $(\mathbf{m l})$ from $\mathbf{5 0 0} \mathbf{~ m l}$ | Thickness | Summary of comments |
| :---: | :---: | :---: | :---: | :---: |
| Method B1 | $7 \pm 1$ | 500 ml | No change | Mild bacon flavour and salt taste |
| Method B2 | $2 \pm 1$ | 500 ml | No change | Strong bacon flavour, |
| Method B3 | $2 \pm 1$ | 500 ml | No change | Strong bacon flavour. <br> overpowered by salt taste |
| Method B4 | $3 \pm 1$ | 500 ml | No change | Little or no bacon flavour or salt |
|  |  |  |  | taste |

## D. 4 Conclusions

Results indicated that the method of boiling the basmati rice in the milk/cream mixture and then straining the rice and retaining the cooking liquid (method R1) was the most appropriate method for transferring the basmati rice flavour to the ice creams. Results indicated that the method of soaking the cooked bacon in the milk/cream mixture and then straining the bacon and retaining the soaking liquid (method B1) was the most appropriate method for transferring the bacon flavour to the ice creams.

## Appendix E:

# Volatile composition table for food 

## pairing study

Table E. 1 Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms.

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Alcohols |  |  |  |  |  |  |  |
| 1-butanol | P | P | NP | P | P | P | P | alcohol |
| 1-heptanol | P | P | P | P | P | P | P | citrus, herbal, woody |
| 1-hexanol | P | P | P | P | P | P | P | grassy |
| 1-octanol | P | P | P | P | P | P | P | fatty, citrus |
| 1-pentanol | P | P | P | P | P | P | P | pungent, bready, winey |
| 1-penten-3-ol | P | P | NP | P | P | P | P | pungent |
| 1-propanol | P | NP | P | P | P | P | P | alcohol |
| isoamyl alcohol | P | P | NP | P | P | P | P |  |
| 2-hexen-1-ol | P | P | NP | P | P | P | P | green fruit, unripe banana |
| Isobutanol | P | P | NP | NP | P | P | P | sweet, musty |
| tert-butanol | P | P | P | P | P | P | P | camphor |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 2-pentanol | P | P | NP | P | P | P | P | winey, ethereal |
| 2-penten-1-ol | P | P | P | P | P | P | P | fruity, plastic |
| 2,3-butanediol | P | P | P | P | P | P | P | cocoa, fruity, buttery |
| 3-hexen-1-ol | P | P | NP | NP | P | P | P | grassy, herbal |
| Prenol | P | P | P | P | P | P | P | fruity, floral |
| 3-octen-1-ol | P | P | P | P | P | P | P | earthy, herbal, grassy |
| 3-methyl, 6-hepten-1-ol | P | NP | NP | NP | P | P | P |  |
| isohexyl alcohol | P | P | P | P | P | P | P | nutty |
| Ethanol | P | P | P | P | P | P | P | ethereal |
| phenylethyl alcohol | P | P | P | P | P | P | P | floral |
| 1,2-butanediol | P | P | P | P | P | P | P | sweet, acetic acid |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 2-butenal |  |  |  | Aldehydes |  |  |  |  |
| 2-heptenal | P | P | P | P | P | P | P |  |
| 2-hexenal | P | P | P | P | P | P | floral |  |
| 2-methyl-butanal | P | P | NP | P | P | P | P | fatty, herbal |
| isobutyraldehyde | P | P | P | P | P | P | P | cocoa, nutty, musty |
| 2-pentenal | P | P | P | P | P | P | P | floral |
| 2, 4-heptadienal | P | P | P | P | NP | P | P | pungent, fruity |
| 3-methyl-2-butenal | P | P | P | P | P | P | P | fatty, oily, cinnamon |
| 3-methyl-butanal | P | P | P | P | P | P | P | sweet, fruity, pungent, nutty |
| acetaldehyde | NP | P | P | P | P | P | P | fruity, fatty, almond |
| fruity, apple, chocolate, musty |  |  |  |  |  |  |  |  |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | $\mathbf{B}+\mathbf{B N}$ | $\mathbf{B + O}$ | $\mathbf{B + R}$ | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| benzeneacetaldehyde | P | P | P | P | P | P | P | pungent, floral, sweet |
| Butanal | P | NP | NP | P | P | P | P | pungent, cocoa, bready, nutty |
| Decanal | P | P | P | P | P | P | P | sweet, floral, citrus |
| Heptanal | P | P | P | P | P | P | P | oily |
| Hexanal | P | P | P | P | P | P | P | grassy, fruity, sweet |
| Methional | P | P | P | P | P | P | P | potato, meaty |
| Nonanal | P | P | P | P | P | P | P | fruity, floral, orange, fatty |
| Octanal | P | P | P | P | P | P | P | fruit, orange, fatty, waxy |
| Pentanal | P | P | P | P | P | P | P | bready, fruity, nutty |


|  | Esters |  |  |  |  |  |  |  |
| :---: | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :---: |
|  | amyl isovalerate | P | NP | NP | NP | P | NP |  |
| 1-methylbutyl butanoate | P | NP | NP | NP | P | P | P |  |
| 1-methyloctyl butanoate | P | NP | NP | NP | P | P | P |  |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | $\mathbf{B + B N}$ | $\mathbf{B + O}$ | $\mathbf{B + R}$ | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 2-methylbutyl butanoate | P | P | P | P | P | P | P | fruity, buttery, fatty |
| 2-methyl cyclohexyl | P | P | NP | NP | P | P | P |  |
| butanoatethyl |  |  |  |  |  |  |  | sweet, fruity, citrus, apple |
| hexyl 2-methylbutanoate | P | NP | NP | NP | P | P | P | fruity, apple, spicy |
| isobutyl propanoate | P | NP | NP | P | P | P | P | rum, pineapple |
| 2-methylbutyl butanoate | P | NP | NP | NP | P | P | P | fruity, buttery, fatty |
| ethyl 2-methyl butanoate | P | P | P | P | P | P | P | fruity, apple |
| Manzanate | P | P | P | P | P | P | P | fruity, pineapple |
| 2-methylbutyl propanoate | P | NP | NP | NP | P | P | P | fruity, pineapple |
| 2-pentenyl butanoate | P | P | P | P | P | P | P |  |
| 3-methylbutyl 2- | P | P | P | P | P | P | P |  |
| methylbutanoate |  |  | P | P | P | P | P | P |
| isoamyl propanoate | P | fruity, apricot, pineapple |  |  |  |  |  |  |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| isoamyl butanoate | P | P | P | P | P | P | P | fruity, banana |
| isoamyl valerate | P | P | P | P | P | P | P | fruity, apple |
| hexyl isovalerate | P | NP | NP | NP | P | P | P | sweet, fruity, apple |
| isobutyl 3-methylbutanoate | P | P | P | P | P | P | P |  |
| isoamyl isovalerate | P | P | P | NP | P | P | P | sweet, fruity, apple |
| isoamyl butyrate | P | NP | NP | NP | P | P | P | fruity, banana |
| butyl butanoate | P | P | P | P | P | P | P | fruity, pineapple |
| butyl pentanoate | P | NP | NP | NP | P | P | P | sweet, fruity, apple |
| butyl propanoate | P | P | P | P | P | P | P | earthy, sweet |
| ethyl butanoate | P | P | P | P | P | P | P | fruity, pineapple |
| ethyl hexanoate | P | P | P | P | P | P | NP | sweet, fruity, banana |
| hexyl butanoate | P | P | P | P | P | P | P | fruity, pineapple |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| isopentyl hexanoate | P | NP | P | P | P | P | P | fruity, banana, apple |
| 2-methylbutanol acetate | P | P | P | P | P | P | P | fruity, banana, apple, sweet |
| methyl butanoate | P | P | P | P | P | P | P | fruity, apples, pineapple |
| propyl butanoate | P | P | NP | P | P | P | P | fruity, sweet, pineapple |
| 2-hexen-1-ol acetate | P | P | P | P | P | P | P | sweet, apple, banana |
| isobutyl acetate | P | P | NP | P | P | P | P | fruity, floral |
| 2,3-octanedione | P | P | P | P | P | P | P | buttery |
| 2-methylbutyl acetate | P | P | P | P | P | P | P | fruity, banana, sweet |
| 2-pentanol acetate | P | P | P | P | P | P | P | fruity, apple, metallic, chemical |
| 2-penten-1-ol acetate | P | P | P | P | P | P | P |  |
| isoamyl acetate | P | P | P | P | P | P | P | Sweet, pear |
| 3-hexen-1-ol acetate | P | P | P | P | P | P | P | sweet, fruity, apple, banana, floral |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| prenyl acetate | P | P | P | P | P | P | P | fruity, banana, sweet, floral |
| 3-octen-1-ol acetate | P | NP | NP | NP | P | P | P | herbal, fruity, oily |
| 4-hexen-1-ol acetate | P | P | NP | NP | P | P | P | fruity, apple, banana, waxy |
| 4-methyl-2-pentyl acetate | P | P | P | P | P | P | P | fruity |
| 4-penten-1-yl acetate | P | P | P | P | P | P | P | fruity |
| butyl acetate | P | P | P | P | P | P | P | fruity, banana |
| ethyl acetate | P | P | P | P | P | P | P | pear, acetone, fruity, sweet |
| hexyl acetate | P | P | P | P | P | P | P | fruity, sweet, fatty, apple |
| methyl acetate | P | P | NP | NP | P | P | P | sweet, fruity |
| pentyl acetate | P | P | NP | NP | P | P | P | banana, apples |
| propyl acetate | P | P | NP | P | P | P | P | fruity, pear |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 2-heptanone |  | P | P | P | P | P | P | P |
| 2-hexanone | P | P | P | P | P | P | P | banana, fruity |
| 2,3 butanedione | P | NP | NP | P | P | P | P | acetone |
| 2-butanone | P | P | P | P | P | P | P | buttery, sweet, caramel |
| 2-pentanone | P | P | P | P | P | P | P | sweet, fruity, wine, banana, wood |
| 3-pentanone | P | P | NP | P | NP | P | P | Acetone |
| 4-hepten-2-one | P | P | NP | NP | P | P | P |  |
| Acetoin | P | P | NP | P | P | P | P | sweet, buttery, fatty |
| Acetone | P | P | P | P | P | P | P | pungent, floral |
| acetyl valeryl | P | NP | NP | NP | P | P | P | buttery, cheesy |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including descriptive terms.

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Sulphur compounds |  |  |  |  |  |  |  |  |
| carbon disulphide | P | P | P | P | P | P | P | sweet ether-like |
| dimethyl sulphide | P | P | P | P | P | P | P | cabbage, sulphurous |
| dimethyl disulphide | P | P | P | P | P | P | P | fetid meat |
| methanethiol | P | P | P | P | P | P | P | decomposing cabbage garlic |
| Phenyls |  |  |  |  |  |  |  |  |
| ethylbenzene | P | P | P | P | P | P | P | sweet, gasoline |
| p-xylene | P | P | P | P | P | P | P | medicinal, solvent |
| Toluene | P | P | P | P | P | P | P | sweet, pungent, benzene-like |
| Terpenes |  |  |  |  |  |  |  |  |
| Limonene | NP | P | P | P | P | P | P | sweet, citrus |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence (P) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | $\mathbf{B}+\mathbf{O}$ | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 4-ethylphenol | Phenol |  |  |  |  |  |  | smoky, woody, medicinal, savoury |
|  | P | P | P | P | P | P | P |  |
| Furan |  |  |  |  |  |  |  | fruity, earthy, metallic |
| 2-pentylfuran | P | P | P | P | P | P | P |  |
| Alkenes |  |  |  |  |  |  |  | pine, earthy, woody |
| Pinene | P | P | P | P | P | P | P |  |
| 1,4-dimethyl Pent-4-enyl | P | P | P | P | P | P | P |  |
| Styrene | P | P | P | P | P | P | P |  |
|  |  |  |  | ines |  |  |  |  |
| trimethyl pyrazine | P | P | P | P | P | P | P | nutty, musty, cocoa, potato |
| 2,5 dimethyl pyrazine | P | P | P | P | P | P | P | chocolate, butter, potato |

Table E. 1 (continued) Volatile compositional analysis of unpaired and paired food samples including presence ( P ) and non-presence (NP) in samples with descriptive terms

| Volatile compound | Banana | Oil | Rice | Bacon | B+BN | B+O | B+R | Descriptive terms |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Carboxylic acids |  |  |  |  |  |  |  |
| acetic acid | P | P | P | P | P | P | P | pungent, vinegar |
| hexanoic acid | P | P | P | P | P | P | P | fatty, cheesy, waxy |

P: compound present in sample.
NP: compound not present in sample.

## Appendix F

# List of publications and conference 

presentations

## List of publications

Traynor, M., Burke, R., Frías, J., Gaston, E. \& Barry-Ryan, C. (2013). Formation and stability of an oil in water emulsion containing lecithin, xanthan gum and sunflower oil. International Food Research. (Accepted for publication June 2013).

Traynor, M., Burke, R, O' Sullivan, M., Hannon, J. \& Barry-Ryan. C. (2013). Sensory and chemical interactions of food pairings (basmati rice, bacon and extra virgin olive oil) with banana. Food Research International. 54 (1). 569-577.

Traynor, M., Burke, R., Brunton, N. \& Barry-Ryan, C. (2012) Response surface methodology guided release of two acetate volatiles from an oil-in-water emulsion. Journal of Culinary Science \& Technology. 10 (3). 223-238.

Burke, R., Danaher, P. \& Traynor, M. (2012). The development of molecular gastronomy as a subject discipline at the Dublin Institute of Technology. Dublin Gastronomy Symposium 2012. Dublin. 05/07/2012.

Valverde, J., Burke, R., \& Traynor, M. (2011). Molecular gastronomy in Ireland. Journal of Culinary Science and Technology. 9 (4). 205-211.

## Conference presentations

Traynor, M., Burke, R. \& Barry-Ryan, C. (2013). Development of novel flavoured ice creams: The who and why of consumer acceptability. Annual Research Chefs Conference. Charlotte. NC. USA. March 6-9, 2013.

Burke, R., Danaher, P. \& Traynor, M. (2012). The development of molecular gastronomy as a subject discipline at Dublin Institute of Technology. Dublin Gastronomy Symposium 2012. Dublin. June 5-6, 2012.

Traynor, M., Burke, R., Brunton, N. \& Barry-Ryan, C. (2011). Effect of Salt Concentration and pH on the release of two banana volatiles from a model emulsion system. Annual Research Chefs Conference. San Antonio. TX. USA. March 20-23, 2012.

Traynor, M., Burke, R., Frías, J., Gaston, E. \& Barry-Ryan. (2011). Influence of soy lecithin, xanthan gum and sunflower oil concentrations on the formation and stability of an oil-inwater emulsion. Annual Research Chefs Conference. Atlanta. GA. USA. March 6-9, 2011.


[^0]:    ${ }^{c}$ centre points.

[^1]:    $\beta_{\mathrm{i}}$ : estimated regression for the main linear effects.
    $\beta_{\mathrm{i}}^{\mathrm{j}}$ : the estimated regression coefficients for the quadratic effects.
    $\beta_{\mathrm{ii}}$ : the estimated regression coefficients for the interaction effects.

    * significant ( $\mathrm{p} \leq 0.05$ ).
    ** highly significant ( $\mathrm{p} \leq 0.01$ ).
    ${ }^{* * *}$ very highly significant $(\mathrm{p} \leq 0.001)$

[^2]:    Samples with the different letters ( $\mathrm{a}-\mathrm{d}$ ) are significantly different ( $\mathrm{p} \leq 0.05$ ).
    $B+R(1-5)$ : banana and basmati rice ice cream formulations.
    *: optimum banana and basmati rice ice cream formulation.

[^3]:    Samples with different letters ( $\mathrm{a}-\mathrm{d}$ ) have significantly different means within each column ( $\mathrm{p} \leq 0.05$ ).
    Panel A: representing the general retail market.
    Panel B: the panel representing the culinary educated market.
    $\mathrm{B}+\mathrm{BN}$ : banana and bacon ice cream.
    $B+R$ : banana and basmati rice ice cream.

