Survey on the Consumption of Cinnamon-Containing Foods and Drinks by the UK Population

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Executive Summary

The main aims of this study were to:

- identify cinnamon-containing foods and drinks commonly consumed in the UK,
- generate new and reliable data on coumarin content of these foods, and
- estimate dietary exposures for adults, children and South Asian groups.

To achieve these objectives, commonly-consumed foods were identified from all major supermarkets, restaurants/takeaways, cafes, ethnic food stores and home-made. A food list containing/labelled with cinnamon and/or mixed spice and/or cassia was sub-divided into 14 food group categories; *bakery products; breakfast cereals; cereal bars; tea/beverages; ice cream/puddings; snack products; rice meals; vegetable dishes; meat dishes; soups/sandwich fillers; cooked/reformed meats; spices; cooking sauces and infant foods*. In all, a list of 80 foods was prioritised using set criteria, and analysed for coumarin using HPLC. A Food Frequency Questionnaire, FFQ, was developed using this food list and a cinnamon survey was conducted in a representative sample of UK population including adults (n= 1011), children (n=162) and South Asians (n=100) using picture cards and portion sizes. Dietary exposures to coumarin were calculated using harmonised approaches, deterministic and probabilistic models.

Coumarin levels (mg/kg for foods; mg/l for beverages) ranged widely; among the foods containing highest levels of coumarin were spices; ground cinnamon (1657mg/kg), mixed spice (456mg/kg) and *garam masala* (124.5 mg/kg in retail samples and 56.5mg/kg in homemade samples). Some curry spices including curry spice, *tandoori, Madras, korma and tikka* contained 52-63.6 mg/kg whilst cinnamon stick contained 86.7mg/kg. Among the infant foods Organix carrot cake contained 11 mg/kg. Several foods were not found to contain coumarin above the detectable level of1mg/kg.

Average dietary exposure to coumarin in the UK population at the medium (50^{th} percentile consumer) percentage of adults was 0.0018 mg/kg-bw/day which was less than the TDI of 0.1mg/kg-bw/day. However, for ethnic groups the exposure was significantly higher (0.022mg/kg-bw/day) and for a small minority of the population surveyed, the dietary exposure exceeded the recommended TDI. Further, follow-up, studies may be required to adequately assess the safety of individuals within this particular sub-group.

There is potential to use the data presented here to compare estimated exposures from this study with the recent NDNS food intake data.

1.0 Introduction

Coumarin (also known as 1,2-benzopyrone, 2H-benzopyran-2-one, o-hydroxycinnamoic acid-δ-lactone, *cis*-o-coumarin acid lactone, coumarinic anhydride and 2-*oxo*-2H-1-benzopyran) is a biologically-active naturally-occurring flavouring found in many plants (including cinnamon, *tonka* beans and sweet clover), essential oils (such as cinnamon bark, cassia leaf and lavender oils) and also, in small amounts, in animal species and edible fruits such as strawberries, cherries and apricots (Lake, 1999). It has a sweet aromatic odour and contributes to the flavour of many foods, beverages and non-food products such as perfumes (Yang *et al.*, 2009). The chemical structure of coumarin is shown in **Figure 1**.

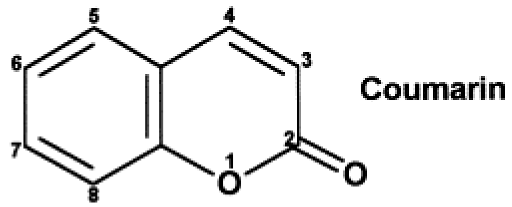

Figure 1 Chemical structure of coumarin (Lake, 1999)

Human exposure to coumarin is mainly through the diet and from its use as a fragrance in bodycare products, perfumes and cosmetics (Felter *et al.*, 2006). The main dietary source of coumarin is cinnamon (Lake, 1999) which could be either cassia cinnamon (*Cinnamomum aromaticum)* with an estimated 3000mg/kg of coumarin or true or Ceylon cinnamon (*Cinnamomum zeylanicum*) which contains about 8mg/kg (Federal Institute for Risk Assessment, BfR, 2006a). Coumarin was widely used as a food flavouring until 1954 when evidence from animal studies prompted toxicological concerns about its use (Clark, 1995). This led to toxicological studies to evaluate the safety of intake by humans and to subsequent regulations on coumarin levels in food. Although animal studies demonstrated liver toxicity (Lake *et al.*, 1989 and 1992; Huwer *et al*., 1991), tumour development (Carlton *et al.,* 1996), and carcinogenic effects (Lungarini *et al.*, 2008) following oral administration of coumarin (Lake *et al.*, 1989 and 1992; Huwer *et al*., 1991), adverse effects after intakes from foods and beverages are rare in humans. Nevertheless, the possibilities of hepatic- and pulmonary toxicities in humans have not been totally excluded but this would depend on coumarin intake levels.

The Scientific Committee on Food (SCF) and the European Food Safety Authority, EFSA, (2004, 2008), therefore, considered risk assessment data obtained by the Federal Institute for Risk Assessment BfR, (2006a, 2006b), Food and Agriculture Organisation/World Health Organisation Expert Committee on Food Additives (Felter *et al*., 2006) as well as scientific studies conducted in humans and animals on the toxicity and metabolism of coumarin (Zhuo *et al*., 1999; Born *et al.*, 2002; Burian *et al*., 2003; Vassallo *et al*., 2004; Lewis *et al*., 2006; Peamkrasatam *et al*., 2006; Satarug *et al*., 2006; Aoki *et al*., 2006; Farinola & Piller, 2007; Rietjens *et al*., 2007, 2008) to set and review a Tolerable Daily Intake (TDI) of coumarin of 0 - 0.1mg coumarin/kg body weight (bw)/day. A TDI of 0.1mg/kg-bw/day would be equivalent to 6 mg/day for an individual weighing 60kg. EFSA recommendedintakes of around 3xTDI for a period of one to two weeks is not of safety concern.

The current **UK Flavouring Regulation**

(http://www.opsi.gov.uk/si/si1992/Uksi_19921971_en_3.htm) states that ***coumarin cannot be added to food as such and specifies coumarin limits for:***

i) *Foods (2 mg/ kg) ,*

ii) *Alcoholic beverages (10 mg/ litre) ,*

iii) *Chewing gum (50mg/ kg) and*

iv) *Caramel confectionery (10mg/kg)*

According to the **European Flavouring Regulation (EC No 1334/2008),** limits will only be set for foods that contribute most to coumarin intake, such as:

i) *Traditional and/or seasonal bakery products containing a reference to cinnamon in the labelling (50 mg/kg),*

ii) *Breakfast cereals including muesli (20 mg/kg),*

iii) *Fine bakery products, excluding traditional and/or seasonal bakery products containing a reference to cinnamon in the labelling (15 mg/kg) and*

iv) *Desserts (5 mg/kg).*

Coumarin is typically found in a range of foods containing cinnamon, such as bakery products, breakfast cereals, spices, confectioneries, desserts and (to a lesser extent) beverages and milk products. EFSA (2006a) calculated an estimated exposure value of 0.025mg coumarin per kg body weight/day, for adults. However, Floc'h *et al*. (2002) reported about human exposures to coumarin of up to 11mg/day from natural food ingredients. Studies by Raters & Matissek (2007) and Sproll *et al*., (2008) reported very high levels of coumarin (from 9mg/kg to 88mg/kg) in cereals, bakery products - especially Christmas puddings -and biscuits and gingerbread biscuits as well as biscuits/cookies which are likely to be cinnamon flavoured.

The Federal Institute for Risk Assessment also highlighted that coumarin exposure through consumption of high amounts of cinnamon-containing dietetic foods for reducing blood sugar in subjects with type 2 diabetes may be of concern (EFSA, 2006b). In the United Kingdom (UK), there is currently very limited information on the levels of coumarin in foods. In the mid-1990s the Ministry of Agriculture, Fisheries and Food (MAFF) conducted a survey on four biologically-active principles, including coumarin, in a range of natural flavouring source materials and preparations. In this survey, high levels of coumarin (up to 157 mg/kg dry weight) were found in some imported cinnamon-based teas. MAFF Survey also suggested that coumarin levels in such teas, ranging from 0.01 to 5.8 mg/kg, would, when consumed exceed the TDI if 3-4 cups were consumed daily (MAFF, 1996). Since this study, there has been a very considerable increase in the range of cinnamon-containing foods and beverages, and changes in the ethnic nature and distribution of the UK population, including increased numbers of ethnic minority groups living in the UK, amongst whom the South Asians are known to use cinnamon in food preparation. Moreover, the results of exposure studies conducted in Germany the last three years, (Sproll *et al*., 2008; Raters & Matissek, 2007) showed appreciable concentrations of the substance in commonly-consumed foods and beverages.

There is also a lack of recent data on coumarin levels in UK foods and beverages, and intake levels, therefore, this project was conducted on cinnamon-containing foods in the UK and on the resulting consumption of coumarin from such foods. Composite samples of prioritised cinnamon-containing foods were analysed by high performance liquid chromatography and human exposures to coumarin estimated by deterministic and probabilistic approaches.

1.1 Objectives

The main objects of the study were to:

1. *prioritise* a list of cinnamon-containing foods,
2. *select* a marketing research company to conduct the food consumption survey,
3. *develop, organise and conduct* a national food consumption survey of cinnamon containing foods,
4. *select* commonly-consumed foods from the survey, *collect and prepare*, food samples for analysis
5. *analyse* samples for coumarin levels, and
6. *submit* final report to FSA following feedback and external review of draft report.

1.2 Justification of the Study

The scientific issue that is addressed is the lack of information on cinnamon-containing foods in the UK and on the resulting consumption of coumarin from such foods. Such data will show whether coumarin intakes in the UK exceed the TDI. In particular, the diets of children and specific ethnic groups such as South Asians will be examined as these groups may be more susceptible to exceeding the TDI.

The data collected on coumarin contents and dietary coumarin exposure from cinnamon-containing foods can be used to:

1. *monitor* levels of coumarin in food and changes in consumption,
2. *identify* the most commonly-consumed cinnamon-containing foods,
3. *provide* new data on the coumarin levels in cinnamon-containing foods,

4. *inform* risk assessments.

1.3 Assessment of Dietary Exposure to Flavouring Substances

To determine whether dietary exposure (the amount of chemical ingested *via* food and drink) to a chemical could cause risk to a population, two major types of information are needed: *food consumption data* and the *concentrations of the chemical in the different food and beverage products* (EFSA, 2005). According to FAO/WHO (2005) and EFSA (2006), dietary data should, ideally, be collected at the individual level (rather than being average data for the population) in order to gather detailed information necessary to provide reliable guidelines to protect the population. The procedure for the collection of dietary data should also take into account non-average, especially high level, consumers who may be at particular risk (FAO/WHO, 2005). Individual dietary data can be collected using various tools, including food frequency questionnaires (quantitative or semi-quantitative), weighed food records and 24-hour food recalls (Kroes *et al.*, 2002). The concentration of the chemical in the food should be determined by reliable and validated analytical methods. However, uncertainties, such as under- and over-reporting of food intake and non-representativeness of the population and foods sampled, could occur (Crispim *et al.*, 2010) in addition to uncertainty associated with the analytical measurement of the chemical concerned (Ellison *et al.*, 2000; Kroes *et al.*, 2002).

In order to assess whether a population is at risk through consuming a chemical in the diet, calculated or estimated exposure levels are compared with a relevant toxicological or nutritional reference value for the dietary chemical of concern (FAO/WHO, 2005). Assessments may be undertaken to determine acute (short-term) or chronic (long-term) exposures, where *acute exposure* cover within a day period and *long-term exposure* covers average daily exposure over a longer period Generally, dietary exposure assessments of potentially toxic food chemicals use default assumptions that tend to over-estimate exposure. The general equation for both acute and chronic dietary exposure is expressed as follows:

$$\textbf{Dietary exposure} = \frac{\Sigma\,(\text{Food chemical concentration} \times \text{Food consumption})}{\text{Body weight}}$$

where, the units usually are milligrams per kilogram body weight (milligrams per kilogram for food chemical concentration; kilograms for food consumption, and kilograms for body weight, FAO/WHO, 2005). Currently, mathematical models are used to measure population dietary exposure levels. These are less expensive, rapid and produce results for large populations, and the results are applicable to past, future or alternative scenarios. Two common models used in dietary exposure assessment are referred to as *deterministic* and *probabilistic*.

A deterministic or point estimate approach is based on a statistic such as the mean or median, or a high percentile of all the consumption values of a considered food in a population of interest. However, this approach does not quantify variability and uncertainties in the input parameters and might lead to over-estimation of intake (WHO, 2005; Rowe, 1986).

A Probabilistic model or approach predicts the probabilities of occurrences of exposures in a population using all available concentration data, taking into account variability and uncertainties in the input parameters (US Environmental Protection Agency, USEPA, 2001; WHO, 2005; Sander and Öberg, 2006).

Furthermore, deterministic models are usually simple to develop and implement but may have limitations, and interpretation of the results can be difficult. Probabilistic models on the other hand give better assessment of dietary exposure than deterministic and require data refinement (FAO/WHO, 2005).

This report describes the assessment of dietary exposure from representative samples of the UK population (children and adults) and South Asians to coumarin by both deterministic and probabilistic approaches or models using individual dietary data (semi-quantitative food frequency questionnaires), actual body weight of individuals and measured concentration of coumarin in selected foods. The probabilistic model utilises a Monte Carlo simulation (Vose, 2001) to explore the range of coumarin exposures, and this takes into account the influence of variability and uncertainty of the data. Monte Carlo simulation is "is a type of simulation that relies on repeated random sampling and statistical analysis to compute the results" (Raychaudhuri, 2008). The FAO/WHO (2005) Guide and Recommendations for Dietary Exposure Assessment of Chemicals in Food were also adhered to.

2.0 Criteria for Identification of Cinnamon-Containing Foods in the UK.

2.1 Sources of Cinnamon-Containing Foods and Beverages

To ensure the data are representative of cinnamon-containing foods and beverages available in the UK and, given that the main source of coumarin in the diet is cinnamon, foods and beverages containing cinnamon were identified (between April, 2009 to March, 2010) from the following sources:

- Popular supermarkets, own brands and manufacturer's

- Ethnic food shops in Leeds, Birmingham and London
- Restaurants
- High street bakeries, cafés and shops selling confectionery
- Internet-based South Asian grocery shops
- A survey of ethnic population in Leeds and London
- Home -made meals and spices (*garam masala*)

2.1.1 Supermarkets

Cinnamon-containing foods and beverages were identified from nine supermarkets [Tesco, Sainsbury, ASDA, Morrisons, The Co-operative (Co-op), Aldi, Lidl, Marks and Spencer, and Waitrose], an internet-based tea shop (Teaworld.co.uk) and Mysupermarket.co.uk (an internet price comparison site and grocery retailer) because of their large market share. In addition, a survey of 4 supermarkets (ASDA, Safeway, Tesco and Sainsbury) was conducted in Leyton, East London (which has a high proportion of residents of South Asian descent) to identify mainstream foods and foods that might be specifically targeted to/purchased by South Asians.

2.1.2 Ethnic food stores

Data were collected from popular ethnic food shops in Leeds (n=2), Birmingham (n=3) and Leyton, East London (n=4). These cities were selected because of popularity of ethnic foods and the high proportion of South Asians living in these cities. Most of the branded ethnic foods distributed in the UK are same therefore, samples from Birmingham, London and Leeds will be the same however, preparation of meals may differ from city to city which is generally influenced by the sub group in that region (Gujarati or Punjabi for instance).

2.1.3 Food manufacturers

Additional information on cinnamon-containing foods and beverages was obtained from Nestle, Cadbury, Kellogg, Jordans, Weetabix Food Company, Dorset Cereal, Eat Natural Limited, Schwartz, Bart, Twinings, Warburtons, Walkers Snack Foods, Pasta King, Loyd Grossman, Mr Kipling, Dominos Pizza Group, Seeds of Change, Weight Watchers, Birds Eye, Sidoli of Shrewsbury, 3663 Food Service, Mars Food Service (Uncle Ben's and Dolmio) and from popular ethnic food manufacturers [Patak, Sharwood's, Natco, Cofresh, East End, Shana, Kashmir Crown Bakeries (KCB) and Rajah].

2.1.4 Restaurants/takeaways

Information on use of cinnamon was also obtained from popular high street restaurants; MacDonalds, Burger King, Kentucky Fried Chicken (KFC), Subway and JD Wetherspoon.

2.1.5 High street bakeries, cafés and confectionery shops

Information on various products that contained cinnamon was obtained from selected high street bakeries (Ainsleys, Greggs, Upper Crust, Delice de France, Bagel Nash, Cooplands and The Pasty Shop), cafés (Starbucks Coffee, Café Nero, Costa, Henry's Café, Café Douwe Egberts and Café Ritazza) and Thornton's Chocolate.

2.1.6 Internet-based South Asian grocery shops

Cinnamon-containing products were also identified from The Asian Cook Shop, Spices of India and Everythingcinnamon.co.uk.

2.1.7 Homemade ethnic foods/spices in Leeds and London

Sixty volunteers of South Asian origin (subjects of Indian, Pakistani and Bangladeshi descent), living in Leeds (n=40) and London (n=20) were interviewed to identify cinnamon-containing home-made foods, beverages and spices.

2.2 Approach

The label and list of ingredients for each food and beverage identified from the above sources were checked for the presence and quantity of cinnamon, mixed spices and/or cassia; the product name and description was also recorded. “Cinnamon” may not be included in the labelling information but may be listed as “cassia” or “mixed spice”. the following approaches were implemented to obtain reliable and validated data.

2.2.1 Web-based searches

Web-based searches of supermarket (Tesco, Sainsbury, ASDA, Teaworld.co.uk and Mysupermarket.co.uk) and manufacturer’s websites were carried out to identify foods and beverages containing cinnamon, mixed spice and cassia from the ingredient list.

2.2.2 Visits to supermarkets, restaurants, cafés and high street bakeries

Where there was little or no information available at the supermarket website, cinnamon-containing foods were personally identified in the store. Visits were requested *via* e-mail and telephone from supermarket head offices and local branch managers. In each case the purpose of the survey, the type of data sought from the stores and the time that the researcher was likely to spend collecting the data was explained. Visits were made to Morrisons, Marks and Spencer, Aldi and Lidl supermarkets, café Nero, Starbuck's café, Costa café, Henry's Café, Café Douwe Egberts, café Ritazza and to Ainsleys, Greggs, Upper Crust, Bagel Nash, Delice de France and The Pasty Shop high street bakeries in Leeds. The staff in these shops was asked which of their pastries or hot/cold beverages contained cinnamon, mixed spice or cassia.

2.2.3 Request for data from supermarket head offices

Additional data were requested by e-mail and telephone calls to Customer Service personnel at the head offices of supermarkets, including The Co-operative (Co-op) and Waitrose (Ocado), restaurants, cafés, high street bakeries, confectioneries as well as manufacturers. Staff was also asked for validation of information obtained on their products on-line and from local branches.

2.2.4 Survey of South Asian population in Leeds and London

Ninety-eight adults [age 30-65 yrs] of South Asian descent and living in Leeds and London were surveyed using a structured and validated questionnaire (**Appendix 1**). The objective was to identify cinnamon-containing foods, both homemade and purchased. These cities were selected because of the population density to recruit a representative sample from sub groups of South Asians living in these particular areas. There was an element of convenience sampling because of our professional and personal contacts to help with such studies in these cities but this did not cause any bias in sampling.

2.2.5 Initial list of cinnamon-containing foods and beverages

The initial list of cinnamon-containing foods from the various outlets and ethnic survey were grouped into bakery, meals, spices, snacks, desserts, sauces etc. and additional information on retail outlets, food descriptor and presence and quantity of cinnamon, mixed spice and cassia [all sources; from the labels, interviews with retail staff and South Asian survey] was compiled into Excel spreadsheets.

2.3 Criteria for prioritisation of cinnamon-containing foods and beverages for inclusion in the national cinnamon survey

Following the initial identification of cinnamon-containing foods and beverages, products were prioritised for inclusion in a Food Frequency Questionnaire (FFQ) for the national cinnamon survey.

These priorities were based on:

1. availability in the four supermarkets with the highest market share in the UK (Tesco, ASDA, Sainsbury and Morrisons[1]).
2. information from the National Diet and Nutrition Survey, NDNS, on adults aged 19-64 years (Volume 5, 2004) and on children between 4-18 years (including the revised consumption data).
3. IGD Report on Average Sales Contribution by Category of Foods and Non-foods from 2006 to 2008[2].

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[1] Source: TNS Worldpanel
[2] Source: IGD Research, 2009

4. IGD Report[2] on Food-to-Go Purchasing Behaviour. This includes cold drinks, snacks, sandwiches, pies, pasties, sausage rolls, biscuits and cakes or pastries.
5. Mintel (2008) keynote reports on cereals, biscuits and cakes

This is to ensure the foods listed were representative of those consumed by the population whilst taking into account seasonal variations in habitual and typical dietary patterns.

2.3.1 Food Frequency Questionnaire (FFQ)

FFQ was selected for the collection of food consumption data because it has been shown to be more accurate and valid than other methods for estimating average dietary exposure to chemicals as well as for absolute non-consumers of certain foods within a specified period (Thompson *et al.*, 2000; Brunner *et al*., 2001; FAO/WHO, 2005). An FFQ including the prioritised cinnamon-containing foods and their typical average portion sizes was designed and pre-tested with 10 volunteers. Typical average portion sizes were based on the serving suggestions indicated by the manufacturer on the packaging and also on information obtained from diet and nutrition surveys and the Food Standards Agency (FSA, 2002). The initial FFQ was revised to make it easier and quicker to administer to the targetted respondents by including coloured picture show cards of the prioritised foods. These cards were pictures of foods as-sold, together with food name and brand, and they helped to increase the reliability of responses and to aid accurate estimation of portion size since responses were based on past intakes. Interview with staff and students (n=10) at the School of Food Science and Nutrition, indicated that the cards were very useful and easy to work out the information being asked. Furthermore, evidence from a study by Crispim *et al*.,

(2010) indicated that subjects may not be clear as to whether the foods they consumed contained cinnamon, hence the use of the picture show cards.

The revised FFQ (**Appendix 2**) consisted of demographic questions (including age, gender, weight, height, ethnicity and socio-economic class), questions about 67 foods and two other questions to identify non-consumers and those who do not remember eating a particular food on the list. Questions relating to home use of cinnamon powder, cinnamon stick and mixed spice (or *garam masala*[3]) were also included. The questions posed relate to whether the foods listed were consumed during the last 7 days and 1 month, the frequency of consumption and the portion size consumed over the last 7 days.

2.4 National Food Consumption Survey

The FFQ was used to survey 1011 adults (aged 16 years and over) and 162 children aged 7-15 years living in the UK. The adult FFQ was adapted for the children's survey by removing questions about spices and sauces (**Appendix 3**). An additional survey was conducted on 100 volunteers of South Asian origin living in Leeds (n=62) and London (n=38).

[3] Blend of roasted and ground spices

2.4.1. Survey type

A marketing company with experience (and ensuring data quality and protection) of bespoke surveys was selected by tender to conduct the survey across the UK. A nationally-representative bespoke approach based on face-to-face interviews was used to reach adults and children across the UK.

2.4.2 Selection of survey respondents

To ensure a representative sampling, a defined 3-stage sampling plan was used to randomly select volunteers across the country. This involved sampling parliamentary constituencies (644 constituents in 11 government office regions and constituencies), and then output areas within the selected constituencies and, finally, selection of individual respondents within the output areas. The 644 parliamentary constituencies in 11 government office regions were classified into 4 urban/rural types (Metropolitan County; other 100% urban; mixed urban/rural; rural) according to socio-economic status. Output areas were randomly selected but with some stratification control based on demographic variables (so-called Mosaic classification) and was checked against the national profile to ensure that it was representative. For each output area, a list of respondents was produced from the most recent UK residential address list (Postal Address File from Royal Mail, UK). Households included in the survey were identified from this list. Twelve individuals were interviewed within each output area. The interviewers used guides, called quota sheets (different for each output area), to determine the type of people to be interviewed based upon their age, gender and employment status..

2.5 Prioritisation of Cinnamon-Containing Foods and Beverages for Coumarin Analysis

Eighty foods and beverages were analysed for coumarin (**Table 1**). Sixty-six foods listed in the FFQ were prioritised for coumarin analysis, in order to:

- *ensure adequate estimation of coumarin exposure from these foods, and*
- *include at least 95% of the population in the exposure estimation.*

The plan was to include all 67 foods in the FFQ but, at the time of the sample collections, *dopiaza* meals were out of stock from the various outlets. An additional 14 samples, including infant foods and home-made *garam masala* spice, were also selected based on the prioritisation criteria described in **Section 2.2**. Home-made *garam masala* was prioritised because the majority of the ethnic population interviewed for the food consumption survey claimed that they make their own using varying amounts of cinnamon; these are to vary in composition to retail samples. The coumarin concentration of home-made *garam masala* was, therefore, used to estimate dietary exposure from this spice for the ethnic population.

2.6 Chemical Analysis of Coumarin in Prioritised Foods and Beverages

2.6.1 Sample collection

Using a defined protocol based on outlet, location, transport and storage, samples were purchased from supermarkets, ethnic foods shops, high street bakeries and cafés. Samples (retail and manufacturer’s brands) of each food and beverage were obtained from Morrisons, Tesco, ASDA and Sainsbury In addition, some foods and spices commonly consumed by the ethnic population were obtained from ethnic shops in Leeds.

2.6.1.1 Samples from supermarkets and ethnic food shops

Supermarkets samples were bought in store and online from the four major food retail outlets in the UK. Depending on the availability of stock at the time of purchase, samples included standard range, luxury and best value brands from each of the supermarkets. Samples of manufacturer's brands, which are sold at these outlets, were also collected. Food samples were also bought from two ethnic shops, CC Continental and Abubakar in Leeds. All purchased samples were transported to the School of Food Science and Nutrition and stored within 30-60 minutes of purchase. Samples purchased on line were delivered to the School in a refrigerated van and stored within 5 to 10 minutes of delivery.

2.6.1.2 Samples from high street bakeries, cafés and restaurants

Pastries and beverages were collected from Bagel Nash, Starbucks café, café Nero, and Costa café in Leeds.

After collection from the various outlets, samples were stored in a cold food storage (-20 ºC and or 5ºC, as required) or room temperature, as appropriate, at the School of Food Science, University of Leeds. The description of the primary samples[4], outlets from which they were purchased and further details are presented in **Appendix 4**.

[4] Primary sample refers to the collection of one or more units initially taken from the total population of the food (Khokhar *et.a.*, 2009).

2.6.2 Sample preparation and recording

Upon arrival at the School of Food Science and Nutrition, all samples were labelled (with permanent marker) with the name of the outlet from which they were purchased and date of purchase recorded in a Sample Collection and Preparation Record Form (**Appendix 4**). Samples were prepared in the Food Processing Laboratory (School of Food Science and Nutrition, University of Leeds) and followed appropriate laboratory practice guidelines. Samples requiring further preparation and cooking were treated according to the preparation instructions on the package. Sample preparation included grilling, cooking/heating in a microwave oven, milling, blending/mixing of composite[5] samples and storage.

2.6.2.1 General laboratory practice

The following laboratory practice guidelines were adhered to:

- checking the list of ingredients of every primary sample for cinnamon, mixed spice or cassia.
- completely thawing all frozen foods before preparation to ensure accurate sample weighing,
- prevention of contamination and loss of samples,
- cleaning (washing and drying) of equipment after preparation of every sample to avoid cross-contamination,
- using clean spoons for drawing of every primary sample to avoid cross-contamination,

[5] Composite sample refers to the mixture formed by combining primary samples (Greenfield & Southgate, 2002, Khokhar *et.al.*, 2009)

- using standard weights to check routinely the accuracy of measuring scales, and
- accurate labelling and appropriate storage of composite samples.

2.6.2.2 Final list of cinnamon-containing foods and beverages

In a few cases, ingredient information from supermarket websites showed products to contain cinnamon, cassia and/or mixed spice, but when purchased the actual labelled information did not show cinnamon, cassia or mixed spice in the ingredient list and therefore, such samples were not included in the composite sample and were removed from the initial list resulting in the final list (**Appendix 5**). This information further emphasises the need for close scrutiny of samples, and the inaccuracy of relying entirely on website information.

2.6.2.3 Sample Preparation

A written record (**Appendix 4**) was kept of all primary samples which included the list of foods, names, brands, date of purchase, expiry date, listed ingredients, quantity or weight of food, state of food as purchased, sample handling and cooking procedures. Some samples required further preparation, such as heating, cooking, grilling and milling, prior to mixing into composite sample.. In each case the manufacturer’s instruction on the food package was followed. All warm or hot samples were allowed to cool before weighing and mixing. Further details of the procedure and utensils used were recorded in the Sample Collection and Preparation Record form.

2.6.3 Preparation of composite sample

A *food group composite approach* was selected for generating coumarin concentration data. Although the individual food approach has a major advantage in enabling the contribution of each food to overall exposure to be determined and, therefore, provides dietary exposure for various segments of the population, large number of samples must be analysed to obtain a reliable representation of foods consumed by the population. The composite sample approach is, therefore, more economical but requires that foods should be carefully selected to reflect national representativeness (including regional and ethical dietary habits), *retail outlets* {supermarkets with the largest market share, ethnic food shops, cafés and bakeries} and *brands* (standard range, luxury, best value and manufacturers brands) as recommended by FAO/WHO (2005) and Khokhar *et. al*., (2009).

The disadvantage of this approach however is the 'dilution effect', whereby the concentration of one or more foods in the composite sample may be significantly in excess of the limit of detection (LOD) or quantification (LOQ) but is diluted to concentrations below the LOD/LOQ.

Samples were collected from various outlets to obtain a representative sample of each type of food or beverage. Depending on the availability of stock at the time of sample collection/purchase, a primary sample included standard range, luxury and best value brands from each of the supermarkets. Samples of manufacturer's own brands, which are also sold at these outlets, were collected across supermarkets. The samples collected from the 4 supermarkets with the largest market share, reflect the variations in the distribution of these foods throughout the UK. Home-made *garam masala* was obtained from 5 volunteers of South Asian origin (Indians and Pakistanis).

Equal weights (edible portions) of primary samples were mixed together using a blender to create a composite sample weighing approximately 400g (**Figure 2**). Composite samples were prepared from primary samples (n=3- 17) which should reflect the variability in the composition due to recipe variation. For each composite sample, two sub-samples were taken (150g each); one to be analysed at the Eurofins laboratory and the other kept in storage at the School of Food Science & Nutrition for future use.

2.6.4 Storage and transport of composite samples

Samples were stored in airtight, plastic screw-cap containers with minimal headspace to avoid moisture loss and contamination. Composite samples were either stored at -20°C (ice cream) or 4°C (all other samples) in the School of Food Science and Nutrition prior to being transported to the Eurofins laboratory.

Samples were transported in polystyrene boxes in a refrigerated van to ensure they remained in their chilled state. Ice cream was transported in a separate polystyrene box in close proximity to dry ice to keep it frozen.

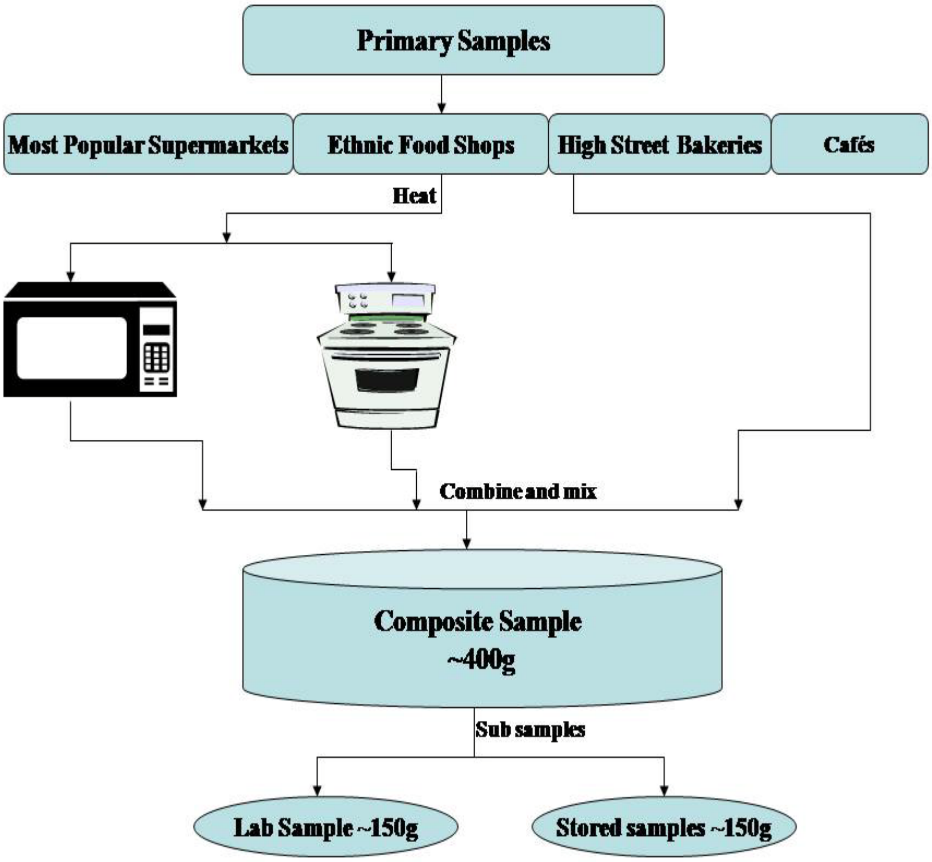

Figure 2. Sample preparation

2.7 Analysis of Coumarin

Coumarin analysis was carried out by the Eurofins Laboratory, Wolverhampton, (UK), which is ISO 17025 accredited and complies with the Joint Code of Practice for Quality Assurance. Eurofins implements quality assurance systems (validation of analytical methods, use of certified materials and internal quality control procedures) and a proficiency testing scheme, which is a legal requirement for laboratories as set by Article 12 of EU Regulation (EC) 882/2004.

Coumarin in food and beverages was determined by high performance liquid chromatography using tandem mass spectrometric detection (HPLC-MS/MS), according to the method of Raters and Matissek (2008). This method is validated, sensitive and allows for selective and fast quantification of coumarin below the maximum tolerance limit of 2mg/kg specified EC Directive 88/388/ECC (European Council, 1988). HPLC-MS/MS is more sensitive (by a factor of 100) compared to standard high-performance liquid chromatography with ultra violet light detection (HPLC-UV), which cannot detect coumarin levels below 2mg/kg. In order to ensure accuracy of the data, analyses were carried out in duplicate on each sample. The limit of quantification (LOQ) was 1.0mg/kg for coumarin in foods and 2.0mg/kg for coumarin in spices.

2.8. Data Evaluation

2.8.1 Evaluation of food consumption data

The food consumption data dataset was compiled in an Excel spreadsheet and meticulously checked for:

i. errors in data entry,

ii. accuracy of data (for example, the number of times a food is consumed in the last 7 days should not be more than the reported frequency of consumption for the past 1 month).

2.8.2 Evaluation of analytical data

The data obtained from Eurofins was inspected for matching codes of samples with their names as in the original list sent to the laboratory. Eurofins also confirmed duplicate analysis of samples. The values were then compared to published values of the same, or similar foods, taking into consideration the number of samples and country of origin, as well as the calculated values based on the ingredient specification (amount of cinnamon, cassia, mixed spice and other spices of known coumarin level) reported by manufacturers/supermarkets.

For similar foods, the coumarin levels obtained were comparable (within average and maximum range) to published values. However, when compared to the ingredients specifications of manufacturers and supermarkets, the calculated values were generally lower that the measured coumarin contents. This could be due to variation in the levels of coumarin in spices.

Adequately defined food consumption survey in a representative sample of the UK population, chemical analysis at accredited laboratory, harmonised food sampling approach, data scrutiny, and laboratory quality control procedures were adopted to produce reliable data by reducing uncertainties.

2.9 Estimation of Dietary Exposure to Coumarin

It was assumed that all the foods analysed that contained cinnamon or related spices which contained coumarin. Thus, for samples with below the limit of quantification (<1mg/kg for foods and 2mg/kg for spices) or non-detected, a value of 1mg/kg was used for foods, 1mg/l for beverages and 2mg/kg for spices. Both **deterministic and probabilistic models** were developed and used to estimate dietary exposures of the population of UK to coumarin. These were based on the reported body weights and estimated food portions of the survey respondents. The exposure for each individual (weight of food consumed multiplied by concentration of coumarin in the food divided by body weight) was determined by calculating and summing the exposure from each food item consumed over 7 days and dividing by 7 to obtain the exposure figure in mg kg^{-1} $bwday^{-1}$; this was repeated for all individuals. When the individual exposures had been estimated these were then ranked in increasing order to allow a cumulative distribution to be constructed.

Furthermore, in order to avoid under-estimation, dietary exposure to coumarin from *dopiaza* meal was based on that of *dopiaza* cooking sauce since this product was out of stock at the time of sample collection and therefore was not analysed. Using the distribution feature of Oracle ® Crystal Ball (Gentry, Blankinship and Wainwright, 2008), which is an add-in for Microsoft Excel Software, the results were used to produce a distribution of exposures of the population by allowing a range of portion sizes as determined from the field analysis of typical portion sizes. Triangular distributions were also used to predict coumarin exposures based on minimum, most likely and maximum food portion sizes. In addition to the average intake forecast following the guidance document of EFSA (2008b), the maximum consumption was also monitored to estimate the highest level of exposure to coumarin.

The model adopted used Monte Carlo simulation approach (Vose, 2001) to explore possible exposures of the population to coumarin, and also addressed variability and uncertainties in the data. The uncertainties addressed were:

- *measurement and sampling uncertainty* of concentration data (repeated food samples taken, LOD quantification max level assigned),
- *sampling uncertainty* of the population surveyed (population size selected)
- *uncertainty of accurately estimating food portion size* (triangular portion size)
- *uncertainty of food intake data* (i.e. recall of past food intake)

*Variability may result from fo*od processing and cooking (e.g. within batch, ingredients, recipe), frequency of consumption of foods listed. This provides a more realistic estimate of the populations' exposure to coumarin especially for individualswho may be at risk.

The Monte Carlo simulation approach relies on repeated random sampling and allows for the quantification of sources of uncertainty and variability contributing toward the exposure estimate. These influences may be correlated with the exposure on the final output (Holmes *et al*., 2005; Raychaudhuri, 2008). Exposure estimates by **Monte Carlo methods may be separated into 2 dimensions**; *the outer loop* quantifies the uncertainty distributions (e.g. uncertain parameters of a concentration data set) and *inner loop* quantifies the sources of variation (e.g. consumption events from individuals). Similarly, 1-dimensional analyses may be conducted whereby only uncertain parameter space is explored. In this study we conducted point estimate exposure assessment and a 1-dimensional Monte Carlo estimation using uncertainty distributions to characterise portion sizes and, in the case of the children, an uncertain distribution for the concentration within the foods.

The Monte Carlo estimates proceed at each iteration by randomly sampling from the uncertain distributions and calculating the average output values from the simulation allowing construction of a cumulative distribution based on a 95% confidence level. This enabled the investigation of the range of coumarin exposures in the population and those at risk of exceeding the TDI. The output data was based on 10,000 random iterations.

2.9.1 Description of model

The model used was based on that of Holmes *et al*., (2005) and the average exposure estimate E_i [*mg/kg-bodyweight/day*] for each respondent and *i* is estimated using the following equation (1);

$$E_i = \frac{1}{7\ BW_i} \sum_{j=1}^{k} f_j c_j p_j \qquad (1)$$

i = 1,2,3,...,N denotes the number of respondents in the survey,

BW_i [*kg*] the body weight of each respondent,

k denotes the number of food components surveyed (adult/ethnic k = 67, children k = 43),

f_j the frequency of consumption of food item j within a 7 day period,

p_j the portion size [kg] of item j,

c_j the concentration [*mg/kg*] of coumarin in food item j. Food items j = 1,2,3,...,k.

The *summation* is divided by 7 to convert the weekly FFQ responses to a daily average. The coumarin concentration in each food item is assigned data values obtained from analysis of 3-X homogenised samples of each food type. (Should a 2-dimensional Monte Carlo study be required the sampling uncertainty could be addressed). Typically, individual or primary samples were selected using different retail outlets, brands and locations portion sizes. These data reflect typical concentrations of coumarin present in the different food types.

Portion sizes may be given variable sizes within a range of typical portions sold. Often a most likely portion size will occur (e.g. 85 grams) but variation does occur.

For probabilistic exposure estimates, Triangular distributions [Minimum portion, Most likely portion, Maximum portion] were used to describe this variation. This approach may account for under- and over-reporting of food intake.

For each individual, an exposure value E_i [mg/kg-bw/day] is calculated, these are then ranked from smallest to largest to give a cumulative distribution. For the quantiles of the distribution several different formulae may be used for symmetrical plotting positions. Such formulae have the form

$$(k - a)/(n + 1 - 2a)$$

for some value of a in the range from 0 to 1/2. The Hazen plotting position, has $a = 0.5$. For large sample sizes, n, there is little difference between the various expressions.

By calculating the exposures for each respondent by summing the contribution from each food component we can identify consumers and non-consumers of dietary coumarin in each of the FFQ. **The dietary exposure to coumarin by the population was based on the following assumptions:**

1. Coumarin is present in foods whenever cinnamon, mixed spices and cassia are present as part of ingredient formulation. This allows variation within the dietary concentration when food components are consumed and further states that cinnamon, mixed spices and cassia cannot be consumed without dietary exposure to coumarin.
2. Exposure to coumarin *via* other routes (e.g. cosmetics) is excluded.

3. Regional and seasonal variation of diets may have appreciable contribution which will require a more targeted study to make an adequate assessment. Similarly, limited time dependency is sampled as the respondents were surveyed over a 5 day period.
4. Brand- and shop loyalty toward consumed products is not specifically addressed. Composite samples comprised several product brands across major retail outlets.
5. Non-detects (<1mg/kg) were assigned the upper value of 1 mg/kg to provide an upper estimate of the exposure and adopt a conservative approach.
6. An average daily exposure is estimated from the responses from a 7-day Food Frequency Questionnaire (FFQ). This may give an underestimate as a 7-day response may be a single consumption event occurring in one day,for example, a respondent may consume 2 cinnamon bagels on a weekend day. This effect may be addressed by conducting a more detailed survey including consumption diaries as in UK National Dietary and Nutrition Surveys (NDNS) where consumed food components are identified, weighed and recorded on a daily basis.
7. Concentration data were taken from the specifically-conducted analysis of composite samples (3-X samples). These are assumed representative average values but further analysis is needed to determine the distribution of sampled data points and, thus, the range of variation typically encountered.
8. Socio-economic groups and associated impacts were not addressed.

3.0 Results

3.1 Demographic Characteristics of Respondents

The majority of respondents from the national food consumption survey (adults aged 16 years and over) were females (n=551, 54.5%), live in the South East region of the UK (**Figure 3)** and were of ideal body mass index, BMI (**Figure 4**, missing values account for total less than 100%). BMI was calculated based on self-reported height and weight of respondents.

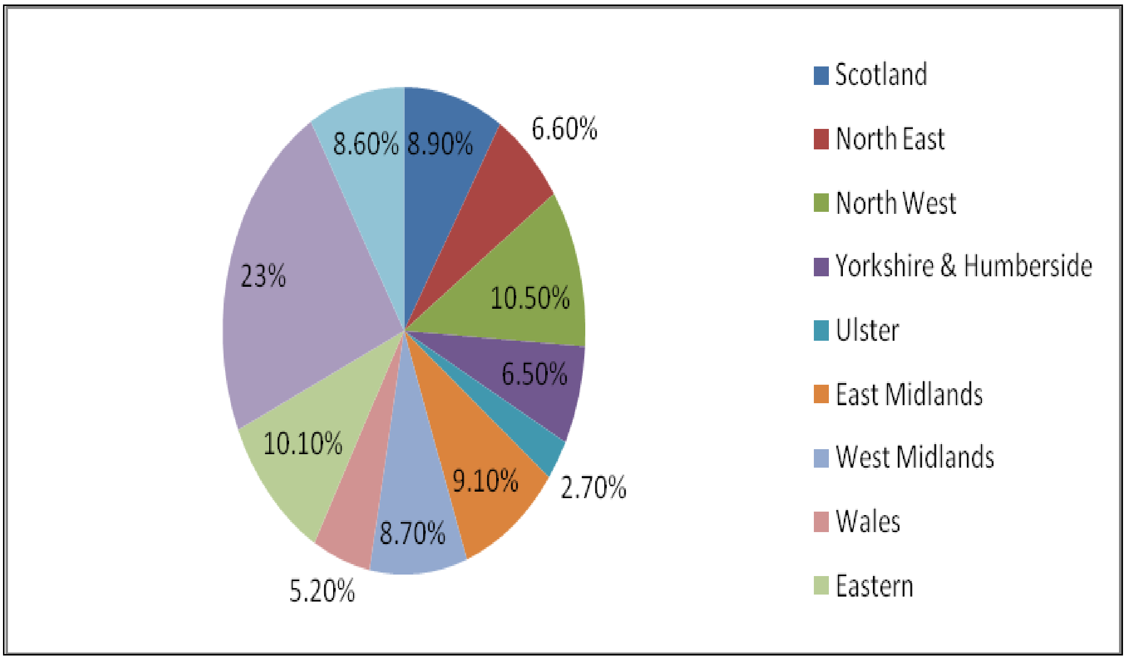

Figure3. Proportion of adult respondents across the UK

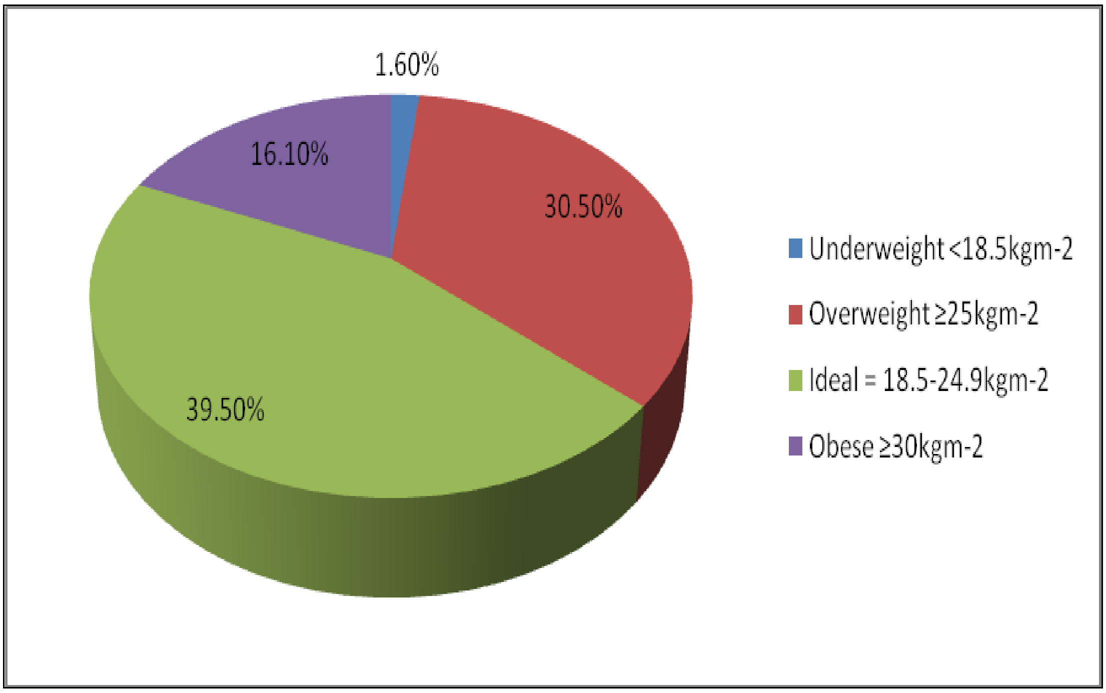

Figure4. Body Mass Index (BMI) of adult respondents

The children [7-11yrs (54.3%), 12-15yrs (45.7%)] included in the survey were from the Midlands (50.6%), South (28.4%) and North (21%) regions of the UK with the majority being girls (n=88, 54.3%). In addition, most of the respondents (16 years and over) for the ethnic food consumption survey were females (78%), of ideal body mass index, 52.5% (overweight = 18.8%, obese = 20.8%, underweight = 3.0%) and of Indian descent, 53.5% (Pakistan = 13.9%, British-South Asian = 30.7%).

3.2 Concentration of Coumarin in Prioritised Foods

New and reliable data on the coumarin content of commonly consumed cinnamon-containing foods were obtained using harmonised, sensitive and validated methods. Cinnamon powder contained the most amount of coumarin followed by mixed spice and retail *garam masala* spice (**Table 1**). The coumarin level of 21 of the 80 foods analysed was below the 1mg/kg limit of quantification. However, all the spices and infant foods analysed contained a measurable amount (>LOQ) of coumarin. Generally, spices contained comparatively higher concentrations of coumarin.

Table 1. Coumarin content of cinnamon-containing foods and drinks consumed in the UK

Composite Sample	No. of Primary Samples	Coumarin (mg/kg)
Bakery		
Fruit Loaf	5	6.2
Cinnamon Bagel	6	21.5
Apple Pie	4	<1
Cinnamon Swirl	4	10.0
Hot Cross Buns	4	10.7
Carrot Cake	10	3.7
Gingerbread Biscuit	4	<1
Naan Bread	7	<1
Oaties- Kelloggs Elevenses oat cookie raisin	4	9.8
Muffins	4	18.2
Chelsea Bun	4	8.95
Fruit Cake	4	3.3
Mince Pie	3	<1
Lotus Original Caramelised Biscuit	4	2.1
Breakfast Cereal/Muesli/Porridge		
Kellogg's Cocopop Mega Munchers	4	<1
Nestlé Curiously Cinnamon	4	<1
Breakfast Cereal – Supermarkets Brand	4	<1
Kellogg's Nature's Pleasure Apple & Blackcurrant Muesli	4	<1
Quaker Oat so simple sweet cinnamon	4	38.2
Cereal Bar		
Cereal Bar	4	<1
Tea/Beverage		
Chai Tea- Twinings *Chai* rich & spicy	4	2.1
Spiced/Herb Tea	6	1.8
Nescafe Cappuccino Powder	4	<1
Cappuccino/Latté Liquid	7	1.4
Chocolate Drink- Nestlé Nesquik chocolate flavour	4	<1

Table 1. Coumarin content of cinnamon-containing foods and drinks consumed in the UK (Contd.)

Composite Sample	No. of Primary Samples	Coumarin (mg/kg)
Ice Cream/Pudding		
Carte D'or Caramel Cinnamon	4	2.0
Ambrosia Rice Pudding with Sultanas and Nutmeg	4	<1
Snack		
Samosa	7	1.8
Onion *Bhaji* (meat/vegetable)	5	<1
Bombay Mix	4	<1
Spring Rolls (meat/vegetable)	4	2.0
Walkers Sensations Barbeque Crisps	4	<1
Roasted Peanuts	3	48.5
Rice		
Pilau Rice (meat/vegetable)	6	1.20
Uncle Ben's Oriental Rice	4	<1
Uncle Ben's Express *Tandoori* Rice	4	<1
Vegetable Dish		
Vegetable Curry	7	3.60
Dhal	4	2.00
Vegetable *Biryani*	4	<1
Meat Dish		
Tikka Masala Meal	14	1.40
Bhuna Meals	4	3.60
Rogan Josh Meals	6	1.85
Jalfrezi Meals	9	<1
Madras meals	11	1.15
Chicken *Korma* Meals	12	<1
Tandoori Meals	4	1.25
Curry Meals	13	<1
Biryani Meals	5	<1
Baked Beans with Sausage	7	<1
Soup/ Sandwich Filler		
Soups	4	<1
Coronation Chicken Sandwich Filler	4	<1
Pickles	4	<1

Table 1. Coumarin content of cinnamon-containing foods and drinks consumed in the UK (Contd.)

Composite Sample	**No. of Primary Samples**	Coumarin (mg/kg)
Cooked Meat		
Quorn Fillets	4	<1
Chicken *Tikka* Slices	5	<1
Sausage	6	<1
Spice		
Mixed Spice	8	456.0
Ground Cinnamon	8	1657.0
Cinnamon Stick	7	86.7
Garam Masala (retail)	6	124.5
Curry Powder	6	51.5
Tandoori, Madras, Korma & Tikka	15	63.6
Biryani Spice	5	18.2
Moroccan Spice	4	63.3
Jerk Spice	4	3.75
Garam Masala (homemade)	5	56.5
Cooking Sauce		
Vindaloo Cooking Sauce	3	6.10
Rogan Josh Cooking Sauce	9	5.10
Barbeque Sauce	3	2.30
Jalfrezi Cooking Sauce	9	2.20
Balti Cooking Sauce	10	3.50
Madras Cooking Sauce	8	4.00
Dopiaza Cooking Sauce	5	4.20
Tikka Masala Coking Sauce	12	2.50
Curry Cooking Sauce	7	<1
Korma Cooking Sauce	15	1.10
Bhuna Cooking Sauce	3	1.60
Heinz Cook at Home Mild Fruity Chicken Curry Sauce	4	<1
Stir-fry Sauce	4	<1
Infant Food		
Heinz Breakfast Oat & Apple cereal for babies	4	5.55
Organix Infant Carrot Cake	4	10.9

3.2 Coumarin levels based on portion size of foods consumed by respondents

The level of coumarin in a portion size was calculated for three portion sizes from the analytical data on the concentration of coumarin. During the interview, respondent estimated his/her food intake based on typical average portion sizes or serving suggestions. Therefore, calculated coumarin content of the foods consumed was based on average, minimum and maximum portion sizes of each item are shown in **Table 2.**

Table 2. Coumarin levels according to the portions size

Food name	**Coumarin concentration (mg/kg)**	**Coumarin[6] concentration in average portion size (mg)**	**Average portion size (g)**	**Minimum portion size (g)**	**Maximum portion size (g)**
Cinnamon bagel	21.5	1.8275	85	85	170
Naan bread	1	0.08	80	60	80
Fruit loaf	6.2	0.217	35	20	70
Carrot cake	3.7	0.13875	37.5	27	50
Fruit cake containing apple and/or spice	3.3	0.198	60	35	60
Gingerbread man biscuit	1	0.01	10	9	15
Lotus original caramelised biscuits	2.1	0.01428	6.8	6.3	7.35
Kellogg's oat cookie raisin	9.8	0.392	40	40	80
Hot cross bun	10.7	0.749	70	70	76
Chelsea bun	8.95	0.6981	78	78	85
Mince pie	1	0.055	55	55	58.3
Cinnamon swirl or cinnamon whirl	10	0.85	85	84.9	85.1
Apple pie	1	0.11	110	78	115
Apple or spiced fruit muffin	18.2	1.4924	82	82	110
Cinnamon favoured breakfast cereal	1	0.045	45	42	50
Cinnamon flavoured muesli	1	0.045	45	45	90
Cereal bar	4.4	0.1892	43	37	50
Samosa	1.8	0.099	55	40	64
Onion *bhaji*	1	0.045	45	35	55
Vegetable/meat spring roll	2	0.12	60	18	60
Cinnamon flavoured potato crisps	1	0.16	160	35	160
Cinnamon flavoured ice-cream	2	0.2	100	100	200
Cinnamon flavoured rice pudding	1	0.212	212	212	425
Herbal/spiced tea containing cinnamon	1.8	0.0036	2	2	4
Chai/masala *chai* tea	2.1	0.00525	2.5	2.5	5
Chai coffee/cinnamon spiced coffee (as served)	1.4	0.364	260	260	350

[6] Food Weight in kg multiplied by Coumarin Concentration in mg/kg = Coumarin Concentration in average portion size (mg per portion)

Table 2. Coumarin levels in portions sizes (contd)

Food name	Coumarin concentration (mg/kg)	Coumarin concentration in average portion size (mg)	Average portion size (g)	Minimum portion size (g)	Maximum portion size (g)
Cinnamon flavoured chocolate drink	1	0.015	15	15	30
Fish/meat/plain/vegetable pilau rice	1	0.25	250	250	340
Biryani rice	1	0.25	250	250	400
Masala dhal	2	0.58	290	150	400
Vegetable curry	3.6	1.2816	356	300	400
Chicken *korma*	1	0.28	280	220	400
Chicken *dopiaza*	4.2	1.5288	364	220	450
Chicken *jalfrezi*	1	0.313	313	196	500
Chicken/beef *madras*	1.15	0.40135	349	168	500
Chicken/lamb rogan josh	1.85	0.592	320	290	350
Chicken/vegetable prawn *tikka masala*	1.4	0.3948	282	220	400
Chicken/prawn/lamb *bhuna*	3.6	1.2888	358	290	500
Tandoori chicken	1.25	0.4875	390	390	475
Chicken/beef/prawn curry	1	0.243	243	144	400
Sausages	1	0.075	75	66.7	150
Chicken *tikka* sliced/chunks/kiev	1	0.198	198	150	260
Quorn fillets	1	0.14	140	140	140
Korma cooking sauce	1.1	0.055	50	35	330
Rogan josh cooking sauce	5.1	0.255	50	35	285
Jalfrezi cooking sauce	2.2	0.11	50	35	285
Bhuna cooking sauce	1.6	0.08	50	35	285
Tikka masala cooking sauce	2.5	0.125	50	35	330
Madras cooking sauce	4	0.2	50	35	285
Balti cooking sauce	3.5	0.175	50	35	250
Dopiaza cooking sauce	4.2	0.21	50	35	250
Barbeque cooking sauce	2.3	0.115	50	35	330
Curry cooking sauce	1	0.05	50	35	250
Vindaloo cooking sauce	6.1	0.305	50	35	250
Cinnamon powder	1657	2.4855	1.5	1.5	6
Cinnamon sticks	86.7	0.30345	3.5	0.88	7
Mixed spices	456	0.684	1.5	1.5	6
Biryani spices	18.2	0.0273	1.5	1.5	6
Garam masala (retail)	124.5	0.18675	1.5	1.5	6
Garam masala (homemade)	56.5	0.08475	1.5	1.5	6
Curry spice/powder	51.5	0.07725	1.5	1.5	6
Korma spice	63.6	0.0954	1.5	1.5	6
Madras spice	63.6	0.0954	1.5	1.5	6
Tikka spice	63.6	0.0954	1.5	1.5	6
Moroccan spice	63.6	0.0954	1.5	1.5	6
Tandoori spice	63.6	0.0954	1.5	1.5	6
Rogan josh spice	63.6	0.0954	1.5	1.5	6
Jerk spice	3.75	0.005625	1.5	1.5	6

3.3 Exposure to Coumarin

Table 3. Number of consumers and non-consumers

Survey group	**Total respondents**	**Consumers**	**Non consumers**	**body weight reported**	**Male consumers**	**Female consumers**
Adult	1011	592	314	105	281	311
Ethnic	100	93	3	4	20	73
Children	162	78	51	33	40	38

After excluding non-consumers and individuals who did not report their body weight, exposure to coumarin was calculated based on 592 adults, 78 children and 93 respondents from the ethnic population as shown in **Table 3**. Given these data, by extrapolating to the population of the UK it can be concluded that about 34.7% (i.e. 314 divided by 906 multiplied by 100) of the population are not exposed to coumarin.

Exposures of dietary coumarin for each consumer were calculated by summing the contribution from each food component identified in the FFQ. Thus for each consumer there may be several contributions e.g. from consumed rice and curry and so an exposure based on the average portion size was initially calculated. These are the results shown in Figure 5 and discussed in section 3.3.1. By dividing the total consumer exposure by their bodyweight provides a standardised measure of exposure per kilogram bodyweight which is more meaningful than a kg/consumer/day since there is variation in consumer bodyweights.

3.3.1 Distributions of exposure to coumarin

The ranked exposure to coumarin based on the deterministic cumulative distribution of the exposure estimates (summed for all foods) for adults, children and ethnic population is presented in **Figure 5**. The medium (50th percentile consumer) percentage of adults (≈0.001800 mg/kg-bw/day) and children (≈0.0015 mg/kg-bw/day) had estimated exposure below the 0.1mg/kg-bw TDI for coumarin. However, for the ethnic population (≈0.022 mg/kg-bw/day) this was significantly higher than for adults and children but below the TDI. Similarly, at the highest levels (97th percentile) of exposure for adults (0.023 mg/kg-bw/day), children (0.012 mg/kg-bw/day) and the ethnic population (0.076 mg/kg-bw/day) the intake of coumarin did not exceed the safety limit. Generally, exposure estimates for children did not exceed the TDI even above the 95th percentile. Thus, the highest exposure of 0.016 mg/kg-bw/day at 99th percentile was below the TDI. However, for adults (≈0.1264 mg/kg-bw/day at 99.9th percentile) and the ethnic population (≈0.1156 mg/kg-bw/day at 99.4th percentile), the high level of exposure exceeded the safety level. A sensitivity test measured by contribution to variance showed that the major source of variation was cinnamon powder (95.3%), followed by *garam masala* (0.75) and cinnamon bagel (0.4%).

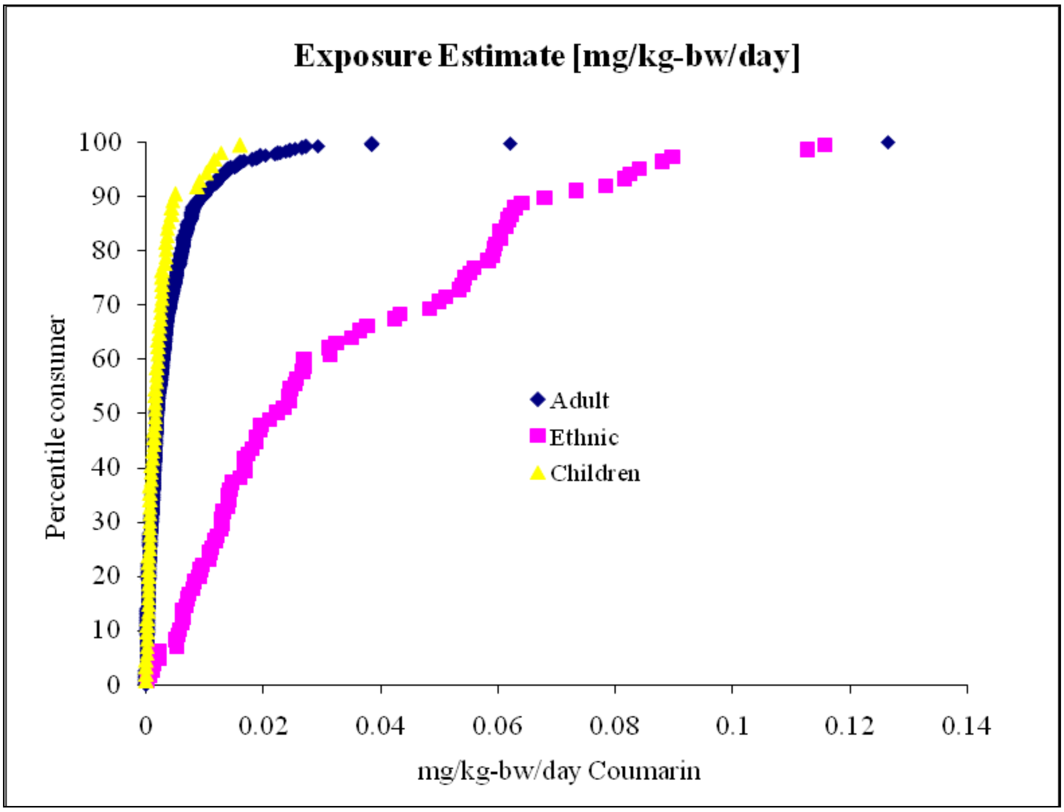

Figure 5. Deterministic cumulative distribution of coumarin exposure estimates for adults, children and ethnic population

Exposure to coumarin was comparable for both males and females with the highest levels being observed in females, as shown in **Figures 6a, 6b and 6c**.

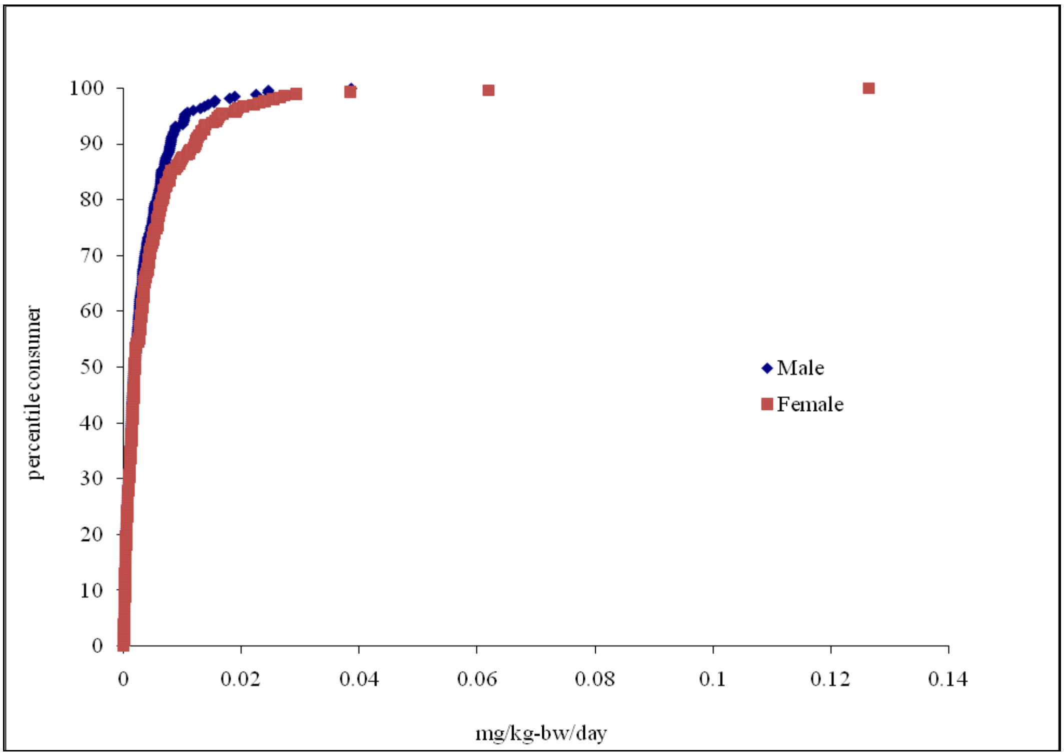

Figure 6a. Comparison of coumarin exposure between adult males and females

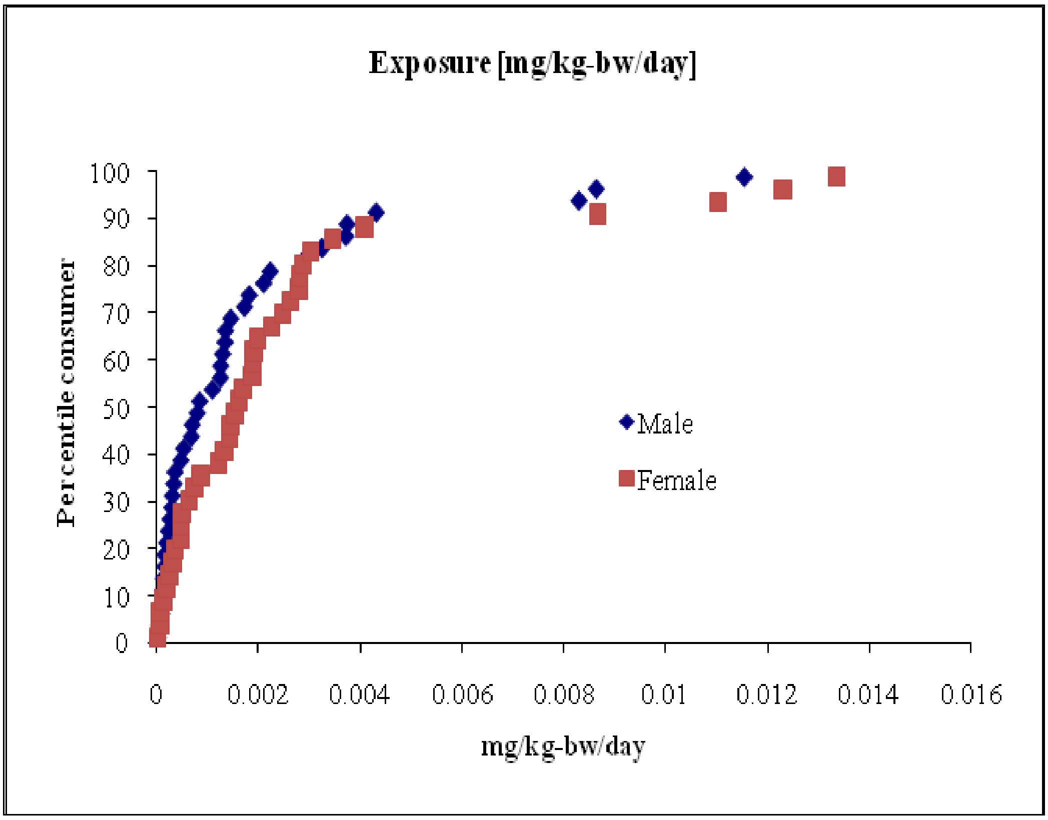

Figure 6b Comparison of coumarin exposure in children by gender

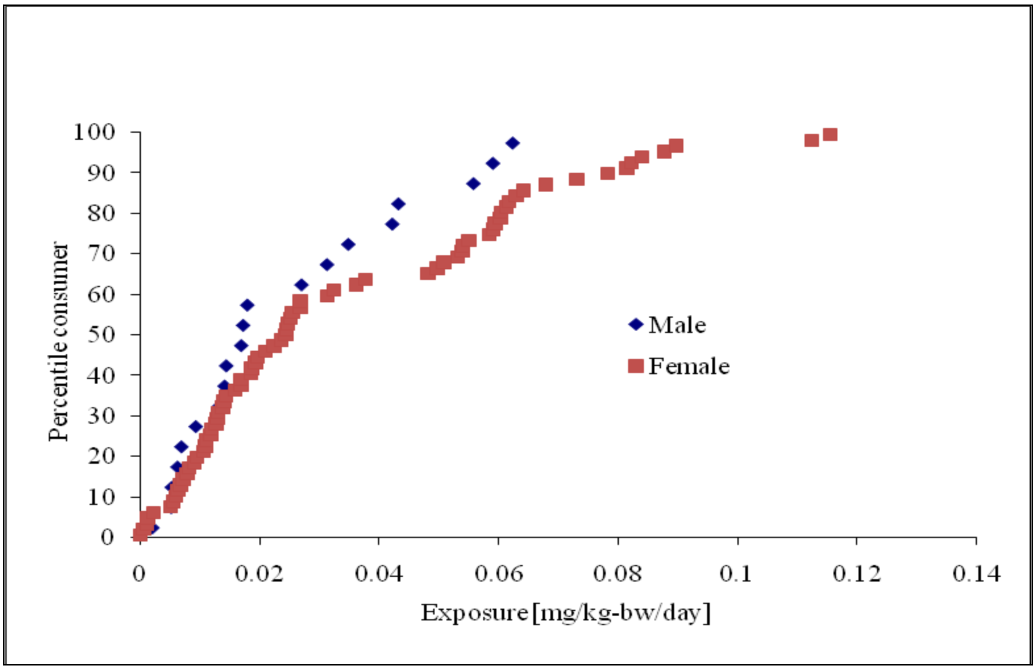

Figure 6c. Comparison of coumarin exposure in adult males and females from the ethnic population

Furthermore, triangular distributions, used to forecasts exposure based on maximum portion or serving sizes of the foods consumed by respondents, are shown in **Figures** 7**a, 7b, 7c and 7d**.

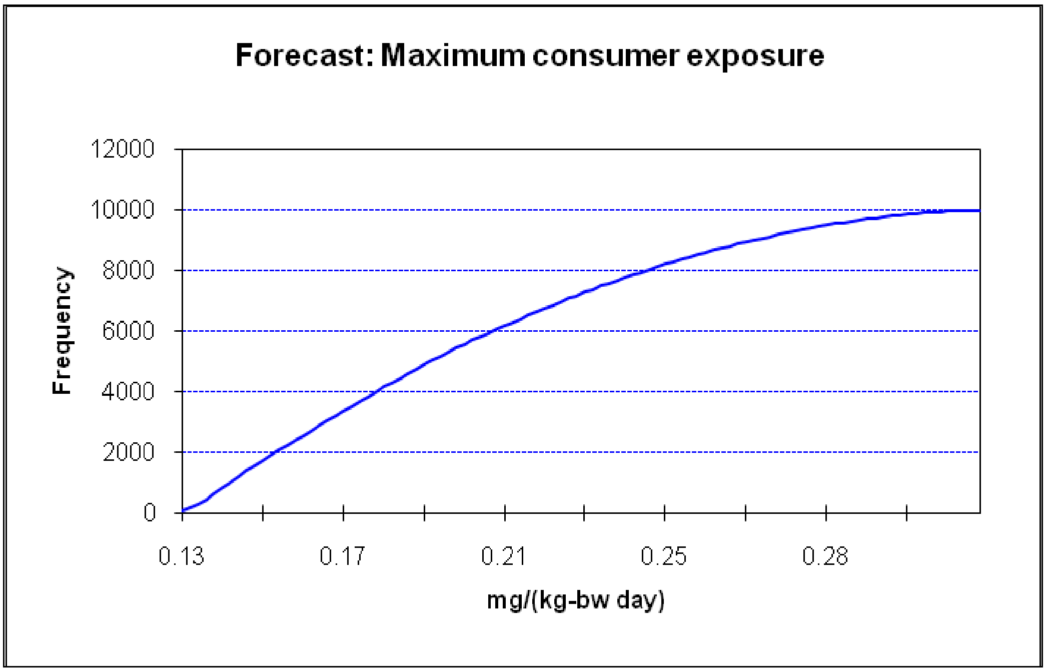

Figure7a. Forecast coumarin exposure for adult maximum consumer

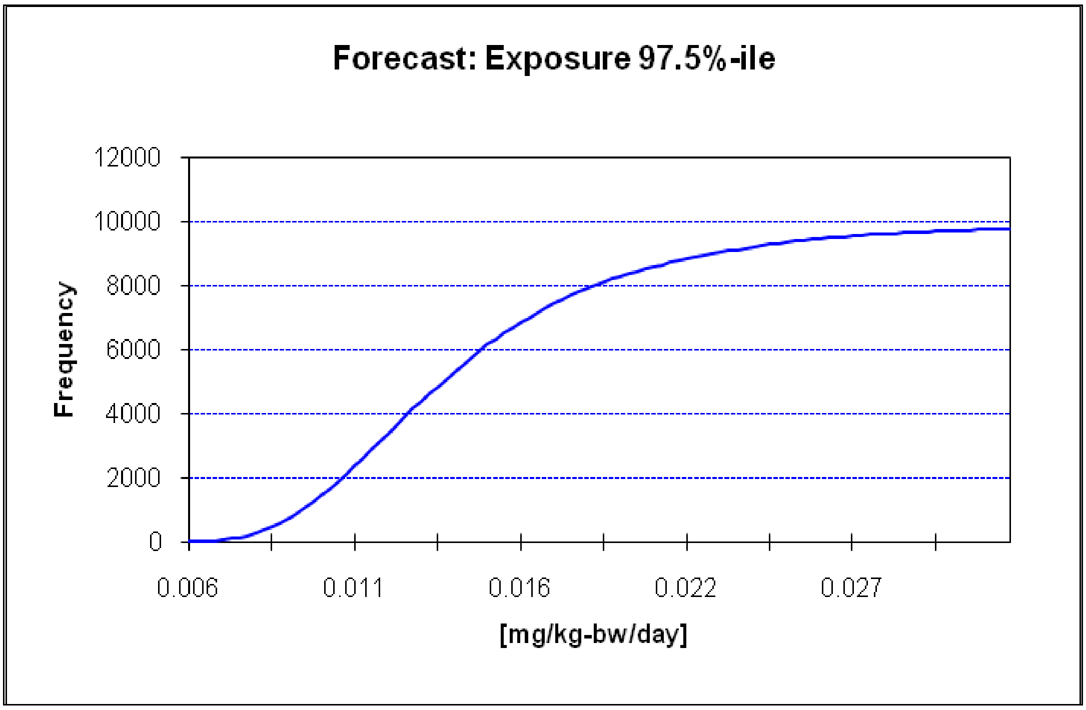

Figure7b. Forecast coumarin exposure for child -maximum consumption

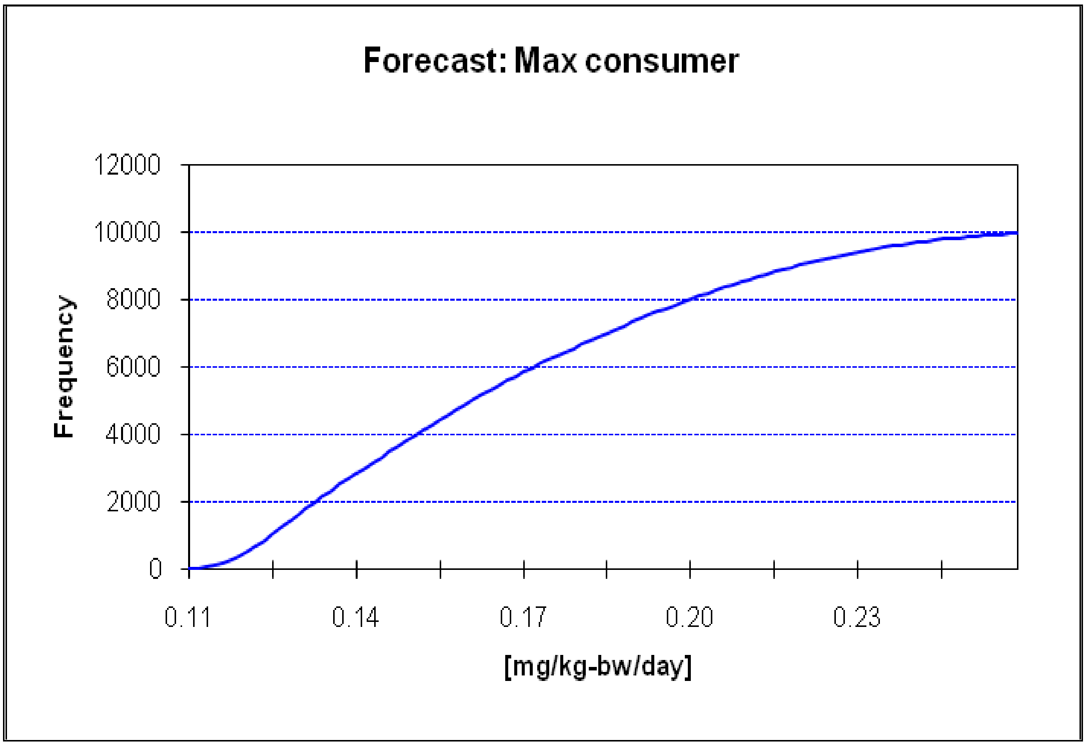

Figure7c. Forecast coumarin exposure for maximum consumption from the ethnic population

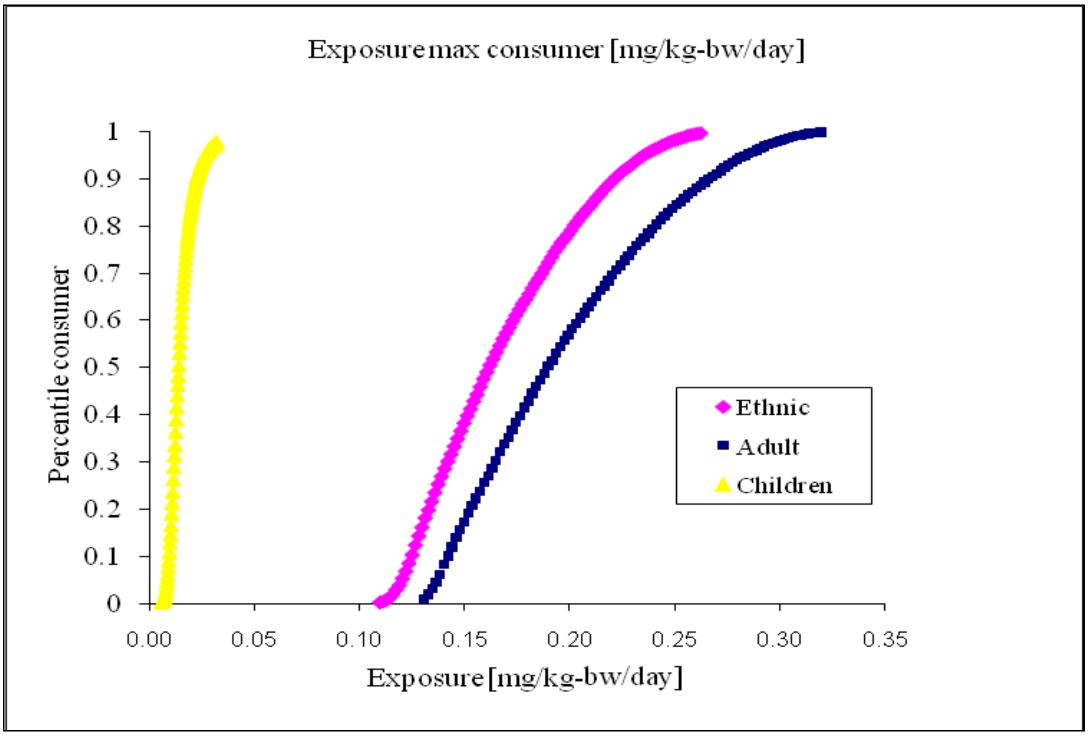

Figure7d. Max consumer overlays for all three surveys

There were 10,000 iterations (in 95% range) but not all data points are displayed in the above Figures.

4.0 Discussion

Dietary exposure to coumarin was estimated using harmonised procedures including deterministic and probabilistic approaches, body weights of respondents and food consumption data of a representative sample (including ethnic population) of the population of UK. Current evidence of coumarin levels in some commonly-consumed foods such as biscuits and pastries suggests intakes by the UK population may exceed the TDI. However, only limited information is available upon which to make a judgement. This study, therefore, provides information on current dietary exposures to coumarin by the UK population. This information will enable the Food Standards Agency to make a risk assessment on coumarin intake by the population. Furthermore, new and reliable data on cinnamon-containing and coumarin concentration in commonly-consumed foods were also obtained.

4.1 Calculated Exposure to Coumarin

The calculated average (median) dietary exposure to coumarin (adults, ethnic population and children) was generally below the safety limit (TDI of 0.1mg/kg-bw). However, at the 99th percentile, exposure for the highest consumers (adults and ethnic population) exceeded the TDI and, therefore, indicating the need for further investigation for subjects in this group who may be at risk. The main contributors (key drivers in exposure levels) to coumarin exposure were cinnamon powder, *garam masala* and cinnamon bagels. Thus, dietary advice targeted at reducing exposure in high level consumers needs to be focused on these food items. This is contrary to the findings of Sproll *et al*. (2008) who estimated that the TDI values could be exceeded by consuming staple products such as bakery products and breakfast cereals, and who suggested the need to regulate coumarin intake. An explorative study by Crispim *et al*. (2010) and based 121 Dutch adults also reported average dietary exposure (mean =

0.07 and median 0.06 mg/kg-bw/day) to coumarin below the TDI but above the safety limit at the 95th percentile (0.12 mg/kg-bw/day). However, unlike the current study, this was not based on a representative sample of the population and uncertainties and variability in the data were not addressed. Furthermore, subject had difficulty correctly identifying cinnamon-containing foods.

Few studies have been conducted to measure human dietary exposure to coumarin. The total human exposure to coumarin, including dietary contributions, has been estimated to be 0.06 mg/kg/day (EFSA, 2004). However, Floc'h *et al*. (2002) reported about 11mg/day human exposure to coumarin from natural food ingredients, which is significantly greater than found in the current study. This may be due to differences in the design of the study, population addressed, types of foods sampled and availability of dietary data. It is necessary to emphasise that, although average dietary exposure of the UK population to coumarin was less than the TDI, when other sources (e.g. body care products and perfumes) are taken into consideration the total value would be greater than estimated. In addition, the data from the current study are likely to underestimate the true exposure to coumarin given that respondents' dietary data was not based on all identified cinnamon-containing foods but only on prioritised ones. However, it would be reasonable to assume the estimates are close to the true value since the FFQ contained commonly-consumed foods identified according to defined criteria.

Given the variation in dietary habits and reported coumarin exposure estimates for different populations, there might a need for countries to harmonise and set safety limit, appropriate for specific sub groups of population.

5.0 Conclusion

New and reliable data on coumarin content of commonly-consumed foods has been generated using validated and harmonised approaches. Highest levels of coumarin were present in ground cinnamon (1657mg/kg), mixed spice (456mg/kg) and *garam masala* (124.5 mg/kg in retail samples and 56.5mg/kg in homemade samples).

A harmonised approach based on deterministic and probabilistic modelling, *realistic food consumption data*, *body weights* of a *representative sample* of the UK population, was developed to reliably estimate dietary exposure to coumarin. The approaches used in this study also addressed sampling and measurement uncertainties.

The average dietary exposure to coumarin by the UK population was less than the TDI of 0.1mg/kg-bw/day and, therefore, did not exceed the safety intake. For a small minority of the population surveyed, dietary exposures exceeded the recommended TDI and, therefore, offer cause for concern. Further studies are necessary to adequately assess the safety of individuals in this group.

The new data on coumarin content in foods may be used by researchers, food manufacturers and legislators to calculate the coumarin content of cinnamon-containing foods and the resulting consumption of coumarin.

6.0 Recommendations

It is recommended that following be further investigated;

- Use of recent National Diet and Nutrition Survey data for the UK may be explored to calculate coumarin exposure.
- While the dietary habits of the UK population continue to change, future exposure studies should be conducted.

- Exposure in high level consumers (although a small number in the present survey) based on more detailed food intake data to identify the key sources or drivers of coumarin in the diet in order to provide adequate advice and intervention.
- Benefits of assessing coumarin exposure by using quantitative FFQ in combination with deterministic and probabilistic models.

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Appendices 1-4