

Retail Surveillance Sampling Programme during Covid-19 pandemic - Method

Sampling

The informal purchasing of samples was carried out by OL staff who were provided with a shopping list and an area of the country in which to shop. All products were purchased at full cost from businesses selling to the general public and the FBOs were not notified that samples were being taken for subsequent testing. The sampling plan focussed on the range of products identified for each sub project and did not target specific businesses.

Geographic Distribution

The surveillance sampling was undertaken in two phases in order to achieve a wide geographic spread. Sampling started in July during Covid-19 restrictions therefore half of the samples were bought from regions local to the laboratories. This was followed by a national sampling plan to cover all of England and Wales when restrictions allowed. A small number of samples were taken in Northern Ireland during regional sampling.

- Phase 1 Regional Sampling commenced July 2020: Sampling was undertaken in the local areas around the OL offices and laboratories including Hampshire, Kent, Lancashire, London, Midlands, Manchester and Northern Ireland providing good regional coverage, whilst remaining compliant with local and national Covid-19 travel restrictions.
- Phase 2 National Sampling (excluding Scotland) commenced August 2020: Areas not covered by the regional sampling were identified and subsequently targeted to ensure a good geographical spread. With Covid-19 restrictions still in place this phase of sampling consisted of both in store and online purchases.

Retail Types

Representative surveillance was also achieved through sampling across a mix of food business operators (FBOs). The project aim was to obtain approximately 25% of samples from large Food Business Operators (FBOs) and 75% from smaller FBOs.

Large FBOs included mainstream supermarkets with national coverage or at least across multiple counties, or large food distributors. Smaller retailers included FBOs smaller than this, such as independent retailers, farm stores, stores operating under franchise, and self-service wholesale stores.

Some samples were purchased via the internet to reflect the consumer move to on-line shopping and provide national coverage during periods of lockdown.

Duplication of Samples

Sampling was coordinated across all five OLs to minimise duplication. Each sampling protocol identified the types of products and these were allocated to the laboratories to purchase with an initial focus on regional or small FBOs. Once purchased samples were added to a central list which was used as a reference for future sampling.

Sample Integrity

In order to ensure that surveillance samples were of a suitable standard for testing sampling protocols were provided for each food commodity/ hazard to ensure that sufficient sample was obtained and that samples were collected, transported and stored under appropriate conditions so as not to adversely impact on the sample integrity or on the quality of the final analytical result. Project protocols are included in Appendix 1 of this report.

Analysis

Each OL holds ISO17025 accreditation and used the most appropriate method for each commodity / hazard analysed using accredited methods if available. All of the surveillance samples in this project were analysed using procedures used for official control samples and the integrity of the samples was maintained at all times with comprehensive records to demonstrate chain of custody.

Raw data for all samples including any replicate analysis, positive and negative controls and quality control materials were recorded and all records kept for a period of at least 12 months. The FSA were provided with photographs of packaging for all unsatisfactory samples as well as the final raw data.

In the event that a laboratory identified something that it considered to be indicate a serious authenticity concern or a significant hazard to human health then the FSA were informed immediately.

P1 Minced and Processed Meat Composition and Speciation

A total of 300 minced and processed meat products were analysed for compositional and speciation compliance. The number of each product type is in the table below.

| Product Type | Number of samples submitted |
|----------------------|-----------------------------|
| Beef Mince | 29 |
| Beef Ready Meal | 30 |
| Beef Burger | 30 |
| Beef Pie | 34 |
| Lamb Mince | 34 |
| Lamb Ready Meal | 30 |
| Lamb Curry / Kebab | 37 |
| Pork Sausages | 31 |
| Pork Mince | 35 |
| Goat Meat / Products | 10 |

Table 1: P1 Sample Numbers by product type

Analysis of meat samples for composition was carried out by five OLs. Proximate analysis was used to measure the amounts of nitrogen, moisture, ash, fat, hydroxyproline and soya protein in the product. Calculations based on the CLITRAVI (Liaison Centre for the Meat Processing Industry in the EU) method for calculation of meat content (9) were used for reporting results.

Analysis of meat samples for speciation was carried out by four OLs. Real-time Polymerase Chain R assays for seven meat species were used to detect and measure (semi-quantitatively) the amount of animal species present. All samples were analysed for the presence of the following meat species:

- Cow (Beef)
- Pig (Pork)
- Sheep (Lamb)
- Goat
- Horse
- Chicken
- Turkey

P2 Fish and Fish Product Speciation

A total of 100 fish and fish product samples were analysed for speciation compliance. The number of each product type is in the table below.

Table 2: P2 Sample Numbers by product type

| Product Type | Number of samples submitted |
|-----------------------------|-----------------------------|
| Cod fillets | 20 |
| Haddock fillets | 20 |
| Plaice fillets | 11 |
| Cod or Haddock fish fingers | 12 |
| Cod or Haddock fishcakes | 11 |
| Named species fish products | 26 |

Speciation analysis was carried out by three OLs using two different DNA techniques.

- PCR-RFLP (using Agilent 2100 Bioanalyzer) of the mitochondrial cytochrome b gene using lab-on-a-chip capillary electrophoresis for end-point analysis enabling accurate sizing of DNA fragments and identification of fish species in raw and cooked foods from reference database.
- DNA extraction and subsequent PCR and sequencing of various, variable mitochondrial DNA regions. Sequences were compared to entries in the public databases NCBI (National Center for Biotechnology Information, USA) and BOLD (Barcode of Life Database).

P3 Spice and Herb Authenticity and Contamination

A total of 375 spice and herb samples were analysed for authenticity. Of the spice samples, 150 were additionally analysed for lead and cadmium levels and 50 were analysed for aflatoxins B1, B2, G1 and G2. The number of each product type is in the table below.

Table 3: P3 Sample Numbers by product type

| Product Type | Number of samples submitted | Metals | Aflatoxins |
|--------------|-----------------------------|--------|------------|
| Turmeric | 50 | 50 | - |
| Oregano | 49 | - | - |
| Thyme | 50 | - | - |
| Black Pepper | 50 | 50 | - |
| Ginger | 50 | 50 | - |

| Product Type | Number of samples submitted | Metals | Aflatoxins |
|--------------|-----------------------------|--------|------------|
| Mixed Herbs | 51 | - | - |
| Spice Mix | 50 | - | 50 |
| Sage | 25 | - | - |

Samples of spices were analysed for authenticity using microscopy by five OLs and one OL tested for metals and aflatoxins. All samples were analysed using light microscopy at both macroscopic and microscopical levels. Observed features were compared with reference literature and reference control authenticated herbs and spices.

Black pepper, ginger and turmeric samples were analysed for cadmium using Flame Atomic Absorption Spectroscopy (AAS) and for lead using a Graphite Furnace AAS.

Spice mixes were analysed for aflatoxins using solvent extraction followed by filtration Immuno Affinity Column clean up (IAC) and HPLC with Kobra cell post column derivatisation.

P4 Basmati Rice and Durum Wheat Authenticity

Samples of basmati rice (40) and durum wheat pasta (25) were analysed for authenticity. The number of each product type is in the table below.

| Table 4: | P4 | Sample | Numbers | by | product | type |
|----------|-----------|--------|---------|----|---------|------|
| | | | | | | |

| Product Type | Number of samples taken |
|-------------------|-------------------------|
| Prepacked Basmati | 20 |
| Cooked Basmati | 20 |
| Prepacked Pasta | 15 |
| Cooked Pasta | 10 |

All basmati rice samples were analysed by one OL and all durum wheat samples were analysed by a different OL. Basmati rice speciation was analysed using PCR & Microsatellite-based DNA analysis using 10 marker microsatellite alleles. The allele pattern was then compared with those of authentic varieties of basmati rice.

Durum wheat speciation was analysed using R-Biopharm DUROTEST SQ Elisa Membrane Kit which uses a monoclonal antibody specific for the protein friabilin which is only present in nondurum wheats.

P5 Undeclared Milk

A total of 140 products which claimed to be free of milk were analysed to establish if the claims were true. The number of each product type is in the table below:

| Table 5: P5 Sample Number | rs by product type |
|---------------------------|--------------------|
|---------------------------|--------------------|

| Product Type | Number of samples taken |
|---|-------------------------|
| Dark Chocolate | 39 |
| 'Free From Milk/Dairy' Chocolate Products | 21 |
| 'Free From Milk/Dairy' Confectionary | 20 |
| Dairy Alternatives: Ice-cream | 10 |
| Dairy Alternatives: Butter | 10 |
| Dairy Alternatives: Milk | 15 |

| Product Type | Number of samples taken |
|----------------------------|-------------------------|
| Dairy Alternatives: Cheese | 15 |
| Dairy Alternatives: Yogurt | 10 |

The products were analysed for the presence of milk by three OLs using two different ELISA kits.

- Neogen Veratox Casein Allergen Quantitative Sandwich Enzyme-Linked Immunosorbent Assay Kit - this kit is used for the quantitative analysis of casein residue in food products and has a limit of detection of 1 ppm casein (NFDM scale).
- RIDASCREEN®FAST Milk this is a sandwich enzyme immunoassay to quantify milk proteins (casein and ß-lactoglobulin) in food containing whey, milk or milk powder and has a limit of detection of 0.3 0.8 ppm milk protein (depending on matrix).

P6 Undeclared Gluten

A total of five samples of gluten-free flour (general all-purpose flour) and five samples of glutenfree flour alternatives (specialist flours such as buckwheat, coconut, gram flour) were selected. Of the ten flours selected, three samples of each were taken from different areas of the country to determine the presence of gluten and any potential variation between batches. The number of each product type is in the table below.

Table 6: P6 Sample Numbers by product type

| Product Type | Number of samples taken |
|-------------------------------------|-------------------------|
| Free From Gluten Flour | 15 |
| Free From Gluten Flour Alternatives | 15 |

Samples were analysed for the presence of gluten by five OLs using two different ELISA kits.

- RIDASCREEN®FAST Gliadin (Art. No. R7002) A sandwich enzyme immunoassay for the quantitative analysis of contaminations by prolamins from wheat (gliadin), rye (secalin), and barley (hordein) in raw products like flours (buckwheat, rice, corn, oats, teff)
- SENSISpec INgezim Gluten R5 ELISA The assay is based on the R5 Monoclonal Antibody, which is specific for proteins from wheat, rye and barley.