3rd Trimester National Microbiological Survey 2004 (04NS3)

European Commission Coordinated Programme for the Official Control of foodstuffs for 2004

Bacteriological and toxicological safety of dried herbs and spices

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Executive Summary

This survey on herbs and spices was undertaken as part of the EU Coordinated Programme for the Official Control of Foodstuffs 2004. The aims of this EU study were to assess:

- the bacteriological safety (*Salmonella* spp., *Bacillus cereus* and *Clostridium perfringens*),
- the *Enterobacteriaceae* levels (as an indicator of irradiation or other treatments) and
- the toxicological safety (aflatoxins) of herbs and spices.

Two additional elements were undertaken in this national study. Their aims were to assess:

- compliance with the labelling requirements for irradiated products and
- the level of heavy metals (lead, cadmium, mercury and arsenic) in herbs and spices.

Sampling for this survey was carried out by Environmental Health Officers (EHOs) at 3 types of premises:

1) Import or Production or Packaging premises or Wholesaler
2) Establishment using large amount of spices for food preparation
3) Retail (including catering).

Single samples were obtained from retail premises, while batch samples (a batch sample comprised of 5 individual samples) were obtained (where possible) from the other premises. Analysis was carried out in the Official Food Microbiology Laboratories (OFMLs) and the Public Analyst Laboratories (PALs).

The following are the main findings:

- 0.9% (6/647) of single samples were classified as unsatisfactory for *Salmonella* spp., while 0.3% of single samples (2/647) and 4% (1/25) of batch samples were classified as unsatisfactory for *B. cereus*. No sample was classified as unsatisfactory for *C. perfringens*.
- A total of 201 samples were tested for irradiation by Pulsed Photo Stimulated Luminescence. Of the 201 samples, 1% (2/201) were found to have been irradiated while a further 4.5% (9/201) showed intermediate or minor signs of irradiation. Information regarding labelling was only provided for one of the two irradiated samples. That sample did not comply with the EU legislative requirements for the labelling of irradiated foods.
- *Enterobacteriaceae* counts of ≤100cfu/g were recorded for 73.9% (478/647) of single samples and 64% (16/25) of batch samples. For the purpose of this programme, the Commission stated that samples with an *Enterobacteriaceae* count ≤100 cfu/g should be suspected of having been irradiated or submitted to similar treatments.
- Spices on the Irish market showed a high degree of compliance with the regulatory limits for aflatoxins. Only 1.6% (2/122) and 0.8% (1/122) of samples failed to comply with the regulatory limits for aflatoxin B1 and total aflatoxins respectively. Levels of the toxic metals arsenic, cadmium, lead and mercury were also generally low, and consumption of these commodities, particularly in the quantities found in a normal diet, is not considered to present any risk to health attributable to the presence of aflatoxins or heavy metals.

1. Introduction

Herbs are leafy parts of soft stemmed plants (e.g. oregano, basil and parsley), while spices are aromatic plant products (e.g. pepper, allspice, cloves, nutmeg, mace, cinnamon, ginger, saffron, and turmeric) [(1)]. Herbs and spices are among the most versatile and widely used food ingredients, their main function being to season, flavour or impart an aroma to foods [(2)]. Demand for herbs and spices has grown in recent years. This has been attributed to a demand for more flavoured foods which are also low in sodium and fat [(3)].

Herbs and spices are important commodities in terms of international trade. They are particularly important to the economies of developing countries (such as India, Egypt, Indonesia, Malaysia, Mexico, Turkey and Brazil) who earn a substantial part of their foreign exchange through export. The major importers are the US, Europe, Japan, East Asian and Middle Eastern countries [(4)]. In 2002, the EU alone imported over 302,000 tons (valued at US$ 744 million) of herbs and spices [(5)]. A scoping study recently undertaken by the Food Safety Authority of Ireland regarding the import of foodstuffs likely to contain specific chemical contaminants, such as spices, has shown that the bulk of Irish spices are imported from another EU country. Only 18% of dried herbs and spices are imported from non EU countries.

As with other agricultural products, herbs and spices are exposed to a wide range of microbial contamination during their cultivation, harvest, processing, storage, distribution and sale. Sources of microbial contamination include the macro environment (i.e. soil or plant in which the products are grown), dust, insects, faecal material and contaminated water [(1,6)]. In addition, microbial contamination can arise from poor handling and poor hygiene practices by food handlers. Drying of herbs and spices (immediately post harvest) is one of the principal factors in controlling microbial growth as it reduces the number of vegetative bacteria present. The remaining flora consists mainly of spore-forming bacteria and moulds, because of their ability to survive on dried materials for long periods [(1,6)] . It should be noted that many herbs and spices are cultivated and harvested in countries with poor sanitary conditions and warm humid climates. These environments increase the potential for microbial contamination and proliferation.

Although herbs and spices are not major contributors to foodborne disease they occasionally contain bacteria that can cause food borne illness (i.e. pathogens) [(1)]. Of particular significance are *Salmonella* spp. and the spore-forming, toxin-producing bacteria *Bacillus cereus* and *Clostridium perfringens*. The presence of pathogens is of concern when herbs and spices are added to ready-to-eat foods (i.e. foods which receive no further processing prior to consumption). In addition, the presence of spore forming bacteria is of particular concern when herbs and spices are added to foods prepared using a cook-chill process. This is because the heat generated during the cooking process will not kill the heat resistant spores but may provide the energy required to initiate spore germination. Subsequent temperature abuse during cooling/storage will result in germination, cell proliferation and possibly toxin production. To date in the European

Union there are no microbiological standards♦ for herbs and spices. However, in Ireland there are microbiological guidelines ϒ for *Ready-To-Eat Foods Sampled at the Point of Sale* [(7)].

Contamination of herbs and spices with moulds also raises concern because under certain conditions some moulds (e.g. *Aspergillus* spp., *Fusarium* spp. and *Penicillium* spp.) are capable of producing secondary toxic metabolites known as aflatoxins. They may be produced either pre- or post-harvest. The latter occurs if crop drying is delayed and if during storage of the crop, water is allowed to exceed the critical values for mould growth. Aflatoxins can lead to deterioration of the liver and kidneys in animals and humans. In addition, some aflatoxins are known carcinogens [(8)]. In the European Union, maximum limits for aflatoxins in *Capsicum* spp. (chilli), *Piper* spp. (pepper), *Myristica fragrans* (nutmeg), *Zingiber officinale* (ginger) and *Curcuma longa* (turmeric) are laid down in Commission Regulation (EC) No 466/2001 [(9)] (amended by Commission Regulation (EC) No 2174/2003 [(10)]).

The control of microorganisms in herbs and spices can be achieved in a number of ways including the implementation of good hygiene practices (environmental, processing, personnel etc). These control measures are addressed in the Codex publication entitled 'A Code of Hygienic Practices for Spices and Dried Aromatic Plants' [(2)]. In addition, irradiation is commonly used as a control measure. Irradiation is a physical treatment where food is exposed to a defined dose of ionizing radiation. It is sometimes referred to as 'cold pasteurisation' since the result achieved is similar to heat based pasteurisation but without the heat [(11)]. Under EU legislation only certain foods can be irradiated; these include dried aromatic herbs and spices (Directive 1999/3/EC [(12)]). Within the EU, irradiated food or food containing an irradiated ingredient must be labelled appropriately, i.e. it must carry the word 'irradiated' in a prominent position either as part of the main label or next to the ingredient that has been irradiated. Currently there are no authorised food irradiation facilities in Ireland and thus any irradiated food marketed within Ireland is imported.

♦ A microbiological standard is a criterion contained in law where compliance is mandatory. The food industry must ensure full compliance with these standards which are monitored by the enforcement agencies.

ϒ A microbiological guideline is a criterion which is not legally enforceable. It provides a benchmark against which unacceptable microbial contamination of food can be identified.

2. Specific Objectives

This survey on herbs and spices was undertaken as part of the EU Coordinated Programme for the Official Control of Foodstuffs 2004. The aims of this EU study were to assess:

- the bacteriological safety (*Salmonella* spp., *Bacillus cereus* and *Clostridium perfringens*),
- the *Enterobacteriaceae* levels (as an indicator of irradiation or other treatments) and
- the toxicological safety (aflatoxins) of herbs and spices.

Two additional elements were undertaken in this national study. Their aims were to assess:

- compliance with the labelling requirements for irradiated products and
- the level of heavy metals (lead, cadmium, mercury and arsenic) in herbs and spices.

3. Methods

3.1 Sample source

Samples were obtained from 3 sources between September and December 2004 (Table 1)

Table 1: Sample source

Sample source	Examples
Import or Production premises or Packaging premises or Wholesaler	Importers, distributors, producers, blenders, packers and wholesalers
Establishments using large amount of spices for food preparation	Bakeries/biscuit makers, soup/sauce/marinade manufacturers, ready-meal manufacturers, snack food manufacturers
Retail premises	Ethnic food shops, food markets, restaurants, supermarkets, grocery shops

3.2 Sample description

The following categories of dried herbs and spices (both pre-packed and loose) were sampled (Table 2):

Table 2: Category of herbs and spices

Category	Example
Capsicum species	Paprika, cayenne, chillies
Piper species	Black, white and green pepper, cubebs
Nutmeg/Ginger/Curcuma	Nutmeg, ginger, curcuma
Other herbs and spices	Garlic, tarragon, parsley, mustard, chives, caraway seeds, sesame seeds, vanilla pods, curry powder

The following were specifically excluded from this survey:

- Fresh herbs & spices
- Herbs & spices with added ingredients such as oil, vinegar, honey, lemon juice, tomato etc. (e.g. herb & spice pastes (e.g. tamarind paste, wasabi paste), fajita mixes, salad dressings, garlic salt, celery salt etc).
- Prepared meals or dishes prepared using herbs and spices.

3.3 Sample size

The size of the sample depended on a number of factors including the sample source, the type of analysis required and the quantity of sample available (Table 3).

Table 3: Details regarding sample size

	Type of analysis	
Sample source	**Microbiological**	**Irradiation/ Aflatoxin/ Heavy metals**
Import or Production or Packaging premises or Wholesaler	Batch sample: 5 x 100g samples♦ (The 5 samples were obtained from the same batch of product)	1kg ✷ ⊗ *See footnote* ▲ *regarding sample size.*
Establishment using large amount of spices for food preparation	Batch sample: 5 x 100g samples♦ (The 5 samples were obtained from the same batch of product)	1kg ⊗ *See footnote* ▲ *regarding sample size.*
Retail (including catering)	Single sample: 100g	250g *See footnote* ▲ *regarding sample size.*

♦ A single sample (i.e. 100g) was obtained if difficulties were encountered obtaining a batch sample.

✷ Where possible sampling was undertaken in accordance with Commission Directive 2002/27/EC and 98/53/EC *(S.I. No. 267 of 2003 – European Communities (Sampling Methods and Methods of Analysis for the Official Control of the Levels of Certain Contaminants in Foodstuffs) Regulations 2003)*. This was undertaken in 1 premises.

⊗ When sampling was not carried out in accordance with Commission Directive 2002/27/EC and 98/53/EC; EHOs were recommended to obtain the 1kg sample by sub-sampling (i.e. 10x100g samples). This was to ensure that the sample was statistically representative of the batch. [*Caution was exercised to ensure that sub-samples of pre-packed products were obtained from the same production batch*].

▲ Smaller sample sizes were obtained if difficulties were encountered obtaining the recommended sample size. A 250g sample allowed for heavy metals, aflatoxin and irradiation testing. A 100g sample allowed for aflatoxin testing only.

3.4 Sampling procedures

1. Generally from each premises, only one sample of each product was submitted per manufacturer (e.g. no more than one sample of Schwartz coriander was submitted from each premises).
2. Loose samples were obtained in a manner similar to that which would be used to serve customers.
3. EHOs completed the relevant questions on the questionnaire at the time of sampling (the questionnaire is outlined in Appendix 1).
4. All samples were stored and transported to the relevant laboratory under dry and ambient conditions.

3.5 Type of Analysis

Samples were submitted to the laboratories for microbiological, irradiation, aflatoxin, and heavy metal testing (Table 4). Microbiological analysis was undertaken in one of the seven Official Food Microbiology Laboratories (OFMLs). Irradiation, aflatoxin and heavy metal testing were undertaken in one of the three Public Analysts Laboratories (PALs). Details of laboratories are outlined in Appendix 2.

Table 4: Information relating to the type of analysis

	Microbiological analysis	**Irradiation/Aflatoxins/Heavy metals**
Parameters to be investigated	• *Salmonella* spp.⋆ • *Bacillus cereus*⋆ • *Clostridium perfringens*⋆ • *Enterobacteriaceae*⋆ ϒ	• Irradiation (by PPSL)⊗ • Aflatoxins (Aflatoxin B1 and Aflatoxin total) ⋆ • Heavy metals (lead, cadmium, mercury, arsenic) ⊗
Relevant laboratory	OFML	PAL

⋆ This analysis was requested by the EC for the EU Coordinated Programme (Commission Recommendation 2004/24/EC [13]).

⊗ Testing for irradiation and heavy metals was not required under the EU Coordinated Programme; however, these studies were undertaken as additional elements in this Irish study.

ϒ *Enterobacteriaceae* testing was carried out as an indicator for possible irradiation or other similar treatments.

3.6 Reporting of results

Laboratory reports (OFMLs and PALs) were forwarded to both the relevant EHO and the FSAI.

3.7 Analysis of results and follow-up/enforcement action

A) Results from microbiological analysis

Upon receipt of the OFML reports, the EHOs determined the microbiological status of the batch and single samples using the criteria outlined in Tables 5 and 6 respectively.

Table 5: Microbiological criteria for batch samples ⁕

Organism	No. of samples in batch	Microbiological status		
		Satisfactory	Acceptable	Unsatisfactory
Salmonella spp.	5	Absent in 25g	N/A	Present in 25g
Bacillus cereus	5	All samples $<10^3$	No more than 1 sample in the range 10^3-10^4	Any sample $>10^4$ or more than 1 sample in the range 10^3-10^4
Clostridium perfringens	5	All samples $<10^2$	No more than 1 sample in the range 10^2-10^3	Any sample $>10^3$ or more than 1 sample in the range 10^2-10^3

Table 6: Microbiological criteria for single samples ⁕

Organism	Microbiological status		
	Satisfactory	Acceptable	Unsatisfactory
Salmonella spp.	Absent in 25g	N/A	Present in 25g
Bacillus cereus	$<10^3$	10^3-10^4	$>10^4$
Clostridium perfringens	$<10^2$	10^2-10^3	$>10^3$

N/A: Not Applicable

⁕ These criteria were proposed by the Commission (Commission Recommendation 2004/24/EC [13]) for the purpose of this survey.

For 'unsatisfactory' samples the EHO determined the type of follow up action to be taken, with advice as necessary from the FSAI or the OFML. Both the microbiological status of the sample and the type of follow up action (where necessary) were recorded on the questionnaire provided.

B) Results from irradiation testing

1) Pulsed Photo Stimulated Luminescence (PPSL)

Testing for irradiation was carried out by Pulsed Photo Stimulated Luminescence (PPSL) in the PALs. Results were reported as negative, minor, intermediate or positive.

2) *Enterobacteriaceae* counts

Enterobacteriaceae results were used as an indicator of irradiation or other similar treatments. For the purpose of this programme, the Commission proposed that samples with an *Enterobacteriaceae* count ≤100 cfu/g (100 cfu/g is the detection limit of the recommended standard ISO 7932:1993 for the enumeration of *Enterobacteriaceae)* should be suspected of having been irradiated or submitted to similar treatments.

According to Directive 2000/13/EC [(14)] (implemented in Ireland by SI No. 483 of 2002 [(15)]) any foodstuff which has been treated with ionising radiation must bear the following information on the label: "irradiated" or "treated with ionising irradiation". Information on labelling was recorded on the questionnaire.

C) Results from aflatoxin testing

Commission Regulation (EC) No 466/2001 [(9)], as amended by Commission Regulation (EC) No 2174/2003 [(10)] lays down the following maximum levels for aflatoxins in *Capsicum* spp, *Piper* spp., *Myristica fragrans* (nutmeg), *Zingiber officinale* (ginger), and *Curcuma longa* (turmeric) (Table 7).

Table 7: Maximum levels for Aflatoxin B1 and Total Aflatoxins in spices (Reg. 466/2001/EC [(9)], as amended by Reg. 2174/2003[10])

Spices	Aflatoxin B_1 (µg/kg)	Aflatoxin B_1+B_2+G_1+G_2 (µg/kg)
Capsicum spp. (dried fruits thereof, whole or round, including chillies, chilli powder, cayenne and paprika)	5.0	10.0
Piper spp. (fruits thereof, including white and black pepper)	5.0	10.0
Myristica fragrans (nutmeg)	5.0	10.0
Zingiber officinale (ginger)	5.0	10.0
Curcuma longa (turmeric)	5.0	10.0

Upon receipt of the PAL reports, the EHO (with advice as necessary from the FSAI) determined suitable action for samples which did not comply with the maximum levels as outlined above. The action taken was recorded on the questionnaire provided. *(Note: Legal action on the basis of these criteria could only be taken on samples obtained in accordance with Commission Directive 98/53/EC (S.I. No. 267 of 2003). No legal action was taken on the basis of results obtained in this survey).*

C) Results from heavy metal testing

There are no legislative limits for heavy metals in dried herbs and spices. The results obtained from this survey were for risk assessment purposes only and therefore no follow-up/enforcement action was required on the basis of the results obtained.

4. Results and Discussion

4.1 Microbiological Results

4.1.1 Overall results

A total of 647 single samples and 25 batch samples were submitted from the 10 health boards for microbiological analysis (Appendix 3).

All samples were analysed for *Salmonella* spp., *B. cereus* and *C. perfringens*. The overall microbiological status of the single and the batch samples are presented in Table 8. 1.2% (8/647) of single samples and 4.0% (1/25) batch samples were classified as unsatisfactory.

Table 8: The overall microbiological status ϒ of single and batch samples

Single / batch samples		**Overall microbiological status** ϒ		
	No. of samples	**Satisfactory (%)**	**Acceptable (%)**	**Unsatisfactory (%)**
Single samples	647	609 (94.1)	30 (4.7)	8▲ (1.2)
Batch samples	25	21 (84.0)	3 (12.0)	1* (4.0)

ϒThe overall microbiological status of the samples was determined as follows:
Satisfactory: Sample (single/batch) satisfactory for all 3 microbiological parameters (i.e. *Salmonella* spp., *B. cereus* and *C. perfringens*)
Acceptable: Sample (single/batch) acceptable for *B. cereus* and/or *C. perfringens* and satisfactory for the remaining parameter(s).
Unsatisfactory: Sample (single/batch) unsatisfactory for 1 or more microbiological parameter and either acceptable or satisfactory for the remaining parameter(s)

▲ 6 single samples were unsatisfactory for *Salmonella* spp. and 2 single samples were unsatisfactory for *B. cereus*. No sample was unsatisfactory for more than 1 parameter.
* 1 batch sample was classified as unsatisfactory for *B. cereus*.

Questionnaires were returned with 341 of the 647 single samples (i.e. a response rate of 52.7%) and with 12 of the 25 batch samples (i.e. a response rate of 48%). The overall microbiological status of samples (single and batch) returned with a questionnaire are presented in Table 9. There is no significant difference (95% confidence interval) between the microbiological status of these samples and the microbiological status of all samples (Table 8). This indicates that samples returned with questionnaires are representative of the total sample population.

Table 9: Overall microbiological status of samples (single and batch) returned with a questionnaire

Single/batch samples	No. of samples	Overall microbiological status[ϒ]		
		Satisfactory (%)	Acceptable (%)	Unsatisfactory (%)
Single	341	327 (96%)	11 (3.2%)	3 (0.8%)
Batch	11	9 (81.8%)	1 (9.1%)	1 (9.1%)

ϒ See Table 8 for details regarding the overall microbiological status

Information recorded on the questionnaire included:

i) **Category of sample** (i.e. '*Capsicum* spp., *Piper* spp., Nutmeg/ginger/curcuma, Other spices & herbs'),

ii) **Type of premises** (i.e. 'Retail', 'Establishment using large amount of spices for food preparation' and 'Import or Production premises or Packaging premises or Wholesaler'),

iii) **Type of packaging** (i.e. loose or pre-packaged) and

iv) **Country of origin** (i.e. the country listed on the label)

Information pertinent to single and batch samples are illustrated in Figures 1 and 2 respectively.

In relation to single samples (Figure 1), all four categories of herbs and spices were sampled in equal proportion. In addition, the majority of samples:

- were obtained from retail premises (72.6%, 244/336),
- were pre-packaged (90.7%, 304/335) and
- originated in Europe (62.2%, 130/209).

The relationship between the overall microbiological status of the single samples and i) the category of sample, ii) the type of premises, iii) the type of packaging and iv) the country of origin is presented in Table 10. The country of origin was the only parameter which had a significant effect (95% confidence interval) on the microbiological status. No sample from Africa or America was categorised as unsatisfactory, while 3.2% and 0.76% of samples from Asia and Europe respectively were categorised as unsatisfactory. However, caution should be taken interpreting this result as it cannot be assumed that the country listed on the label was always the country of origin (e.g. it may have been the country of packing or the country of transport).

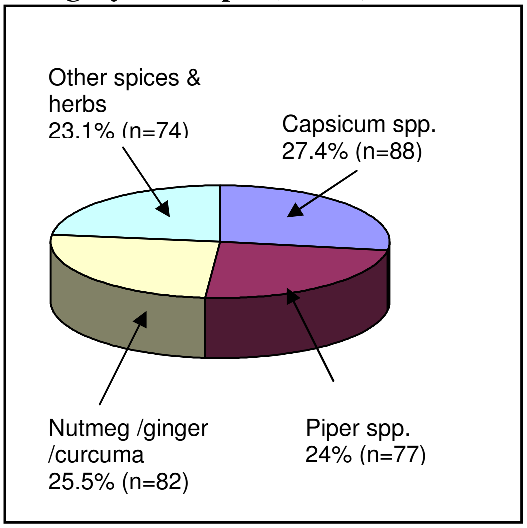
In relation to batch samples,

- The majority were categorised as '*Other herbs & spices*' (72.7%, 8/11).
- The majority of samples were obtained from '*Import or Production or Packaging premises or Wholesaler*' premises (81.8%, 9/11).
- Both 'pre-packaged' and 'loose' herbs and spices were sampled in almost equal proportions and
- Almost half the samples originated in Europe (44.5%, 4/9).

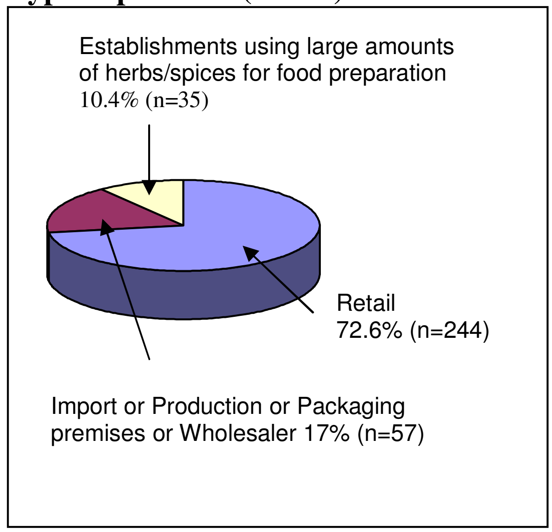
The relationship between the overall microbiological status of the batch samples and each parameter is presented in Table 11. No parameter had a significant effect (95% confidence limit) on microbiological status.

Figure 1: Information recorded on the questionnaire for single samples (n=341)

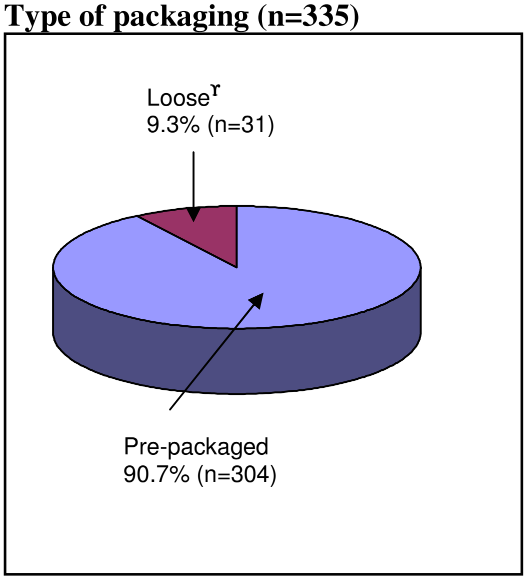
Category of sample (n=321)



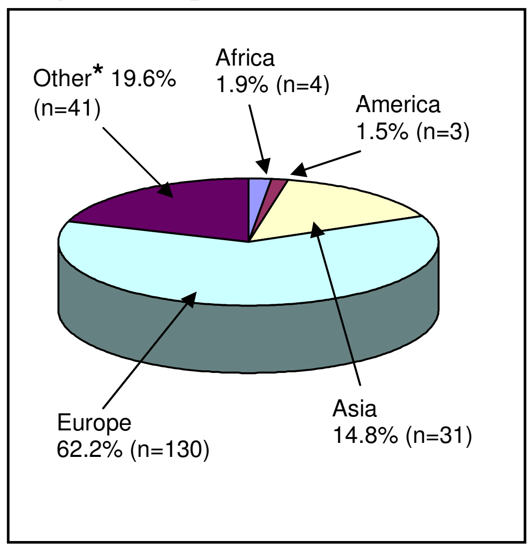
Type of premises (n=336)



Type of packaging (n=335)



Origin of sample[▲] (n=209)



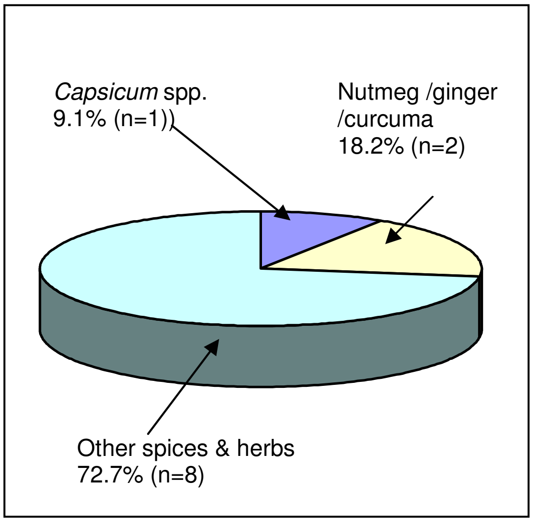
[ϒ] Information on access of the public to loose samples was provided for 29 of the 31 samples (i.e. 93.5%). The public did not have access to 27 of these samples.

[▲] This was the country listed on the label. Caution should be taken interpreting these findings as it cannot be assumed that the country listed on the label was always the country of origin (e.g. it may have been the country of packing or transport). Analysis was carried out by continent rather than country.

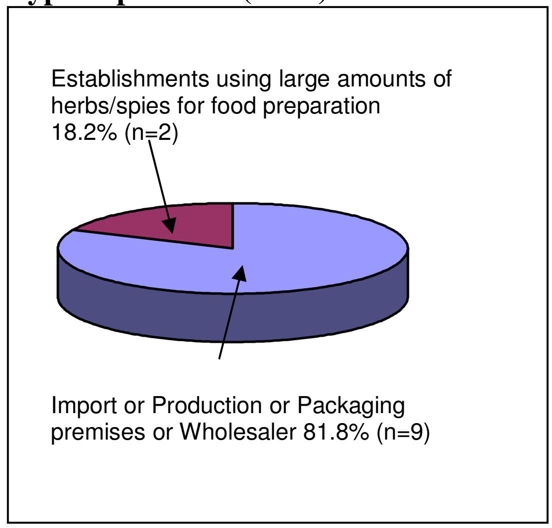
* **Other:** Includes non-EU, packed in Europe, product of more than one country.

Figure 2: Information recorded on the questionnaire for batch samples (n=11)

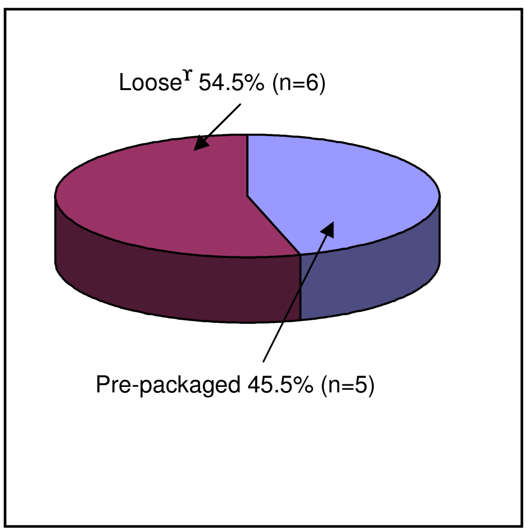
Category of sample (n=11)



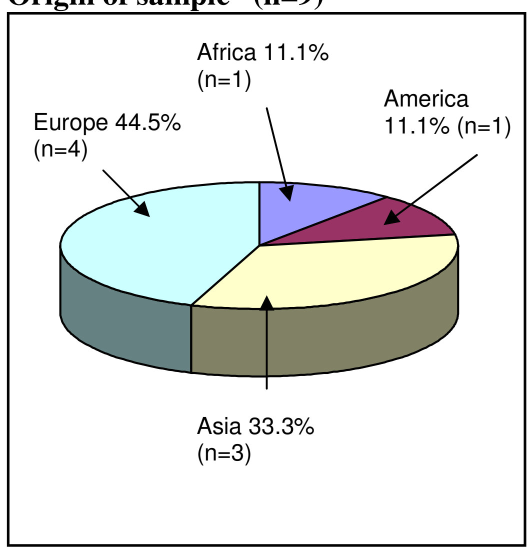
Type of premises (n=11)



Type of packaging (n=11)



Origin of sample▲ (n=9)



ϒ The public did not have access to any loose sample.

▲ This was the country listed on the label. Caution should be taken interpreting these findings as it cannot be assumed that the country listed on the label was always the country of origin (e.g. it may have been the country of packing or transport). Analysis was carried out by continent rather than country.

Table 10: Effect of various parameters on overall microbiological status of single samples

Parameter	Parameter details	Overall microbiological status			Total
		No. of samples satisfactory	No. of samples acceptable	No. of samples unsatisfactory	
Category of sample	*Capsicum* spp.	84	4	0	88
	Piper spp.	77	0	0	77
	Nutmeg /ginger /curcuma	78	3	1	82
	Other spices & herbs	69	3	2	74
	Total	**308**	**10**	**3**	**321**
Type of premises	Retail	233	8	3	244
	Import or Production or Packaging premises or Wholesaler	54	3	0	57
	Establishments using large amounts of herbs/spices for food preparation	35	0	0	35
	Total	**322**	**11**	**3**	**336**
Type of packaging	Pre-packaged	291	10	3	304
	Loose	30	1	0	31
	Total	**321**	**11**	**3**	**335**
Origin of sample	Africa	4	0	0	4
	America	3	0	0	3
	Asia	30	0	1	31
	Europe	128	1	1	130
	Other	36	5	0	41
	Total	**201**	**6**	**2**	**209**

Table 11: Effect of various parameters on overall microbiological status of batch samples

Parameter	Parameter details	Overall microbiological status			Total
		No. of samples satisfactory	No. of samples acceptable	No. of samples unsatisfactory	
Category of sample	*Capsicum* spp.	1	0	0	1
	Piper spp.	0	0	0	0
	Nutmeg /ginger /curcuma	1	0	1	2
	Other spices & herbs	7	1	0	8
	Total	**9**	**1**	**1**	**11**
Type of premises	Retail	0	0	0	0
	Import or Production or Packaging premises or Wholesaler	8	1	0	9
	Establishments using large amounts of herbs/spices for food preparation	1	0	1	2
	Total	**9**	**1**	**1**	**11**
Type of packaging	Pre-packaged	5	0	0	5
	Loose	4	1	1	6
	Total	**9**	**1**	**1**	**11**
Origin of sample	Africa	1	0	0	1
	America	1	0	0	1
	Asia	2	1	0	3
	Europe	4	0	0	4
	Total	**8**	**1**	**0**	**9**

4.1.2 Results by organism

A) *Salmonella* spp.

Salmonellae are pathogens which are one of the most common causes of bacterial food poisoning. Salmonellosis (i.e. the disease caused by *Salmonella enterica*) is principally a food borne disease, although other possible routes of transmission include contact with infected animals or their faecal material, person to person spread and nosocomial infection. In Ireland, 486 clinical isolates of *S. enterica* were referred to the National Salmonella Reference Laboratory in 2003 (crude incidence rate of 11.5 cases per 100,000 population) [(16)].

Salmonellae reside in the intestinal tract of infected animals and humans and are shed in the faeces. Foods subject to faecal contamination (e.g. agricultural products, meat) are among those which have been implicated as vehicles in the transmission of this pathogen to humans [(17)]. Contamination of agricultural products (e.g. herbs and spices) can occur at any stage during cultivation, harvest or post harvest.

Epidemiological studies have shown that outbreaks of salmonellosis have been associated with the consumption of contaminated herbs and spices. Outbreaks have been associated with *S*. Oranienburg in black peppercorns in Norway [(18)], *S*. Weltevreden in black and white peppercorns in Newfoundland [(19)] and *Salmonella* spp. in paprika and paprika products in Germany [(20)]. The presence of salmonellae is of particular concern in herbs and spices when these commodities are added in sufficient quantities to foods that are consumed raw or when added to cooked foods.

In this study 99.1% (641/647) of single samples and 100% (n=25) of batch samples were classified as satisfactory for *Salmonella* spp. (in others words salmonellae were not detected in these samples), while 0.9% (6/647) of single samples were classified as unsatisfactory (i.e. salmonellae were detected in these samples) (Table 12). Details of the samples submitted from each health board in relation to their salmonellae results are outlined in Appendix 4.

Table 12: Microbiological status of samples based on salmonellae results

Single / Batch	Total no. of samples	Microbiological status	
		No. of samples satisfactory (%)	No. of samples unsatisfactory (%)
Single	647	641 (99.1)	6 (0.9)
Batch	25	25 (100)	0 (0)

Information relevant to the 6 unsatisfactory samples is outlined in Table 13.

Table 13: Details of single samples unsatisfactory for salmonellae (n=6)

Sample Description	Category of herbs & spice	Type of premises	Type of packaging	Period remaining on shelf life of product ⊗	Follow up action
Sesame seeds	Other herbs & spices	Retail	Pre-packed	7 months	A verbal warning was issued and a repeat sample was taken. Salmonellae were not detected in the repeat sample.
Turmeric	Nutmeg/ ginger/ curcuma	Retail	Pre-packed	16 months	A verbal warning was issued. A repeat sample could not be obtained as the retailer ceased trading this product.
Curry powder	Other herbs & spices	Establishments using large amounts of herbs/spices for food preparation	Pre-packed	6 months	A repeat sample was taken and *S.* Seftenberg was detected. A written warning was issued.
Extra hot chili pepper	*Capsicum* spp.	Retail	Loose ϒ	16 months	*S.* Bredeney was detected in the sample. 3 repeat samples were taken. Salmonellae were not detected in the repeat samples.
Dried chilli powder	*Capsicum* spp.	Retail	Pre-packed	18 months	3 repeat samples taken (2 formal and 1 informal). Salmonellae were not detected in the repeat samples.
Turmeric	Nutmeg/ ginger/ curcuma	Import/production /wholesaler	N/S *	N/S *	*S.* Agona detected. A written warning was issued and a repeat sample was taken. Salmonellae were not detected in the repeat sample.

⊗ This was calculated to the nearest month

ϒ The public did not have access to this sample

* No questionnaire was returned for this sample

N/S: Not stated

It is worth noting that the period remaining on the shelf life of these unsatisfactory samples ranged from 6 to 18 months. The presence of salmonellae is of particular concern in products with a long shelf life, as improper storage conditions (e.g. temperature and humidity) can lead to multiplication of bacterial cells.

The presence of salmonellae in herbs and spices has been investigated in other studies (some of the studies in which salmonellae were detected are outlined in Table 14). In the studies presented the prevalence of salmonellae in herbs and spices ranged from 0.6% to 6.5%. In this study, the prevalence of salmonellae in 0.9% of single samples is at the lower end of this range.

Table 14: Incidence of salmonellae in herbs and spices

Location of study	Year of study	Sample type	No. of samples tested	Number of samples (%)	
				Salmonellae present	Salmonellae absent
Australia (adapted from 21)	1986✱	Various spices	523	10 (1.9%) ϒ	513 (98.1%)
USA [22]	1987	Imported spices	31	2 (6.5%) ⊗	29 (93.5%)
Austria [23]	1994✱	Herbs and spices	160	1 (0.6%)♦	159 (99.4%)
Ireland (this study)	2005	Herbs and spices	647 single samples 25 batch samples	6 (0.9%)▲ 0 (0%)	641 (91.1%) 125 (100)

✱ This is the publication year, the survey period was not stated

ϒ *Salmonella* spp. detected in black peppercorns (n=8), white peppercorns (n=1) and Indian fungreek seed (n=1). Serotypes isolated: *S.* Binza, *S.* Mbandaka, *S.* Lexington, *S.* Augustenborg, *S.* Alachua, *S.* Mguloni, *S.* Weltevreden
⊗ *Salmonella* spp. detected in 2 samples of black pepper corn (from Indonesia and Brazil). 1 serotype was detected in the sample from Indonesia (*S.* Infantis), while 3 serotypes were isolated in the sample from Brazil (i.e. *S.* Give, *S.* Albany and *S.* Dusseldorf).
♦ *S.* Arizonae was detected in 1 sample of black pepper.
▲ Salmonllae serovars identified in 2 samples: *S.* Bredeney and *S.* Agona

B) *Bacillus cereus*

B. cereus is a spore forming, toxin producing bacterium. It is ubiquitous in nature and is readily isolated from soil, dust and vegetation. Therefore, it is not surprising to find this organism on virtually every raw agricultural commodity [(24)].

B. cereus causes two types of food borne disease:
1) An emetic intoxication due to the ingestion of a toxin preformed in the food (the toxin is extremely stable[ϒ] and cannot be inactivated by reheating) and
2) A diarrhoeal infection due to the ingestion of bacterial cells/spores which produce enterotoxin in the small intestine.
B. cereus counts of 10^5 to 10^6 cells or spores/g of food are usually required to cause illness; however, in some cases lower counts have been implicated [(25)].

Almost all types of foods have been associated with *B. cereus* food poisoning. However, the majority of cases have been linked to heat treated foods which have been subjected to temperature abuse [(25)]. Temperature abuse can result in spore germination and multiplication of the vegetative cells, leading to hazardous levels of bacteria or toxins in the food at the time of consumption. Herbs and spices are common ingredients in heat treated foods and therefore have the potential to contribute to *B. cereus* food poisoning.

In this study *B. cereus* was detected at levels $>10^4$ cfu/g (i.e. at unsatisfactory) levels in 0.3% (2/647) of single samples and in 4% (1/25) of batch samples (Table 15). Details of the samples submitted from each health board in relation to their *B. cereus* results are outlined in Appendix 5.

Table 15: Microbiological status of samples based on *B. cereus* results

Single / batch	Total no. of samples	Microbiological status		
		No. of samples satisfactory (%)	No. of samples acceptable (%)	No. of samples unsatisfactory (%)
Single	647	621 (96.0)	24 (3.7)	2 (0.3)
Batch	25	21 (84)	3 (12)	1 (4)

Details of the samples categorised as unsatisfactory for *B. cereus* are outlined in Table 16.

[ϒ] stable at 126°C for 90 minutes [(26)]

Table 16: Details of samples unsatisfactory for *B. cereus*

Sample	Sample description	Category of herbs & spices	Type of premises	Type of packaging	Follow-up action
Batch⊗	Ginger	Nutmeg/ginger/ curcuma	Establishments using large amounts of herbs & spice for food preparation	Looseϒ	Written warning
Single▲	Curry	Other herbs & spices	Retail	Pre-packed	Verbal warning
Single▲	Garlic powder	Other herbs & spices	Retail	Pre-packed	A repeat sample was taken and the results were unsatisfactory. The batch of product was subsequently destroyed.

ϒ The public did not have access to this sample.
⊗ This sample had a further 19 months remaining on its shelf life.
▲ Information on shelf life was not submitted for these samples.

In other studies on the microbiological status of herbs and spices, *B. cereus* was detected at levels $>10^4$ cfu/g in up to 6.5% of samples (Table 17).

Table 17: ***B. cereus* levels in herbs and spices**

Location of study	Year of study	Sample type	Total no. of samples	*B. cereus* count		
				No. of samples $<10^3$ cfu/g (%)	No. of samples 10^3-10^4 cfu/g (%)	No. of samples $>10^4$ cfu/g (%)
Australia (adapted from 21)	1986 ✹	Various spices	520	330 (63.5)	156 (30)	34 (6.5)
Italy (27)	1996	Various spices	200	192 (96)	4 (2)	4 (2)
Hungary (28)	1988✹	Ground paprika and ground pepper	63	57 (90.5)	5 (8)	1 (1.5)
The Netherlands (29)	1996✹	Herbs/Spices/ Seasonings	6	Range 10^2 - 10^6		
The Netherlands (30)	1985✹	Spices and herbs	100	77 (77)	20 (20)	3 (3)
US (31)	1976✹	Processed spices	110▲	95 (86.4)	15 (13.6)	0 (0)
Ireland (this study)	2005	Herbs and spices	647 (single samples) 25 (Batch samples)	621 (96.0) 21 (84)	24 (3.7) 3 (12)	2 (0.3) 1 (4)

✹ This is the publication year, the survey period was not stated
▲ 89% (88/99) of the isolates tested produced an enterotoxin

C) *Clostridium perfringens*

C. perfringens is a spore forming bacterium. In terms of its oxygen requirements it is one of the less fastidious of the clostridia species and is capable of growing under conditions that are not strictly anaerobic.

C. perfringens is ubiquitous in the environment (soil, dust, vegetation) and is part of the normal intestinal tract of humans and animals. It is present in many raw foods (e.g. herbs and spices, vegetables, meats) as a result of contamination with soil or faecal matter [(32)].

Its ability to grow in food is determined mainly by the storage temperature [(33)]. In cooked foods, low numbers of *C. perfringens* spores often survive the cooking process. Subsequent temperature abuse (during cooling and storage) can result in spore germination and cell multiplication. Ingestion of large numbers of vegetative cells can cause illness (infective dose 10^8cfu/g [(33)]). In the stomach, the vegetative cells which survive the acidic conditions sporulate and release an enterotoxin which causes symptoms including abdominal pain, nausea and acute diarrhoea.

In relation to herbs and spices, the European Food Safety Authority's (EFSA) Scientific Panel on Biological Hazards [(33)] has stated that there is no evidence that adding substantial amounts of herbs and spices to cooked foods poses a real risk for *C. perfringens* induced illness. However, there is a risk if the cooked food is subjected to temperature abuse.

In this study *C. perfringens* was detected at satisfactory and acceptable levels in 98.6% (638/647) and 1.4% (9/647) of single samples respectively. All batch samples (n=25) were categorised as satisfactory (Table 18). Details of the samples submitted from each health board in relation to their *C. perfringens* results are outlined in Appendix 6.

Table 18: Microbiological status of samples based on *C. perfringens* results

Single/Batch sample	Total no. of samples	Microbiological status		
		No. of samples satisfactory (%)	No. of samples acceptable (%)	No. of samples unsatisfactory (%)
Single	647	638 (98.6)	9 (1.4)	0 (0)
Batch	25	25 (100)	0 (0)	0 (0)

In other studies the percentage of samples with unsatisfactory levels (i.e. >10^3 cfu/g) of *C. perfringens* ranges from 1.8 to 7.6% (Table 19).

Table 19: *C. perfringens* levels in herbs and spices

Location of study	Year of study	Sample type	Total no. of samples	*C. perfingens* count		
				No. of samples $<10^2$ cfu/g (%)	No. of samples 10^2-10^3 cfu/g (%)	No. of samples $>10^3$ cfu/g (%)
Australia (adapted from 21)	1986⁕	Various spices	516	406 (78.7)	71 (13.7)	39 (7.6)
US (34)	1975⁕	Processed spices	114	101 (88.6)	11 (9.6)	2 (1.8)
The Netherlands (30)	1985⁕	Spices and herbs	100	41 (41)	57 (57)	2 (2)
Ireland (this study)	2005	Spices and herbs	647 (single samples) 25 (batch samples)	638 (98.6) 25 (100)	9 (1.4) 0 (0)	0 (0) 0 (0)

⁕ This is the publication year; the survey period was not stated

4.2 Irradiation Results

4.2.1 Results from Pulsed Photo Stimulated Luminescence (PPSL) testing

A total of 201 samples were tested for irradiation by PPSL. Overall, 1% (2/201) of samples were found to have been irradiated while a further 4.5% (9/201) showed intermediate or minor signs of irradiation (Table 20).

Table 20: Results from irradiation testing (n=201)

Result	Number of samples (%)
Positive	2 (1)[ϒ]
Intermediate	8 (4.0)
Minor	1 (0.5)
Negative	190 (94.5)
Total	**201 (100)**

[ϒ] Both samples were also tested (by Thermoluminescence) by the *Scottish Universities Research and Reactor Centre* (SURRC).

4.2.1.1 Compliance with labelling legislation in relation to irradiation

Under EU legislation, irradiated food or food containing an irradiated ingredient must be labelled appropriately, i.e. it must carry the word 'irradiated' in a prominent position either as part of the main label or next to the ingredient that has been irradiated. In addition some labels voluntarily carry the wording 'has not been irradiated'.

In this study labelling data was provided for 37.8% (76/201) of the samples which were tested for irradiation by PPSL. A correlation between the labelling data and the results from the irradiation testing by PPSL is provided in Table 21.

Table 21: Correlation between labelling information and results from irradiation testing

Information recorded on the label of the product	No. of samples	Results from irradiation testing by PPSL		
		No. of samples positive (%)	No. of samples intermediate (%)	No. of samples negative (%)
Has been irradiated	3	0	0	3
Has not been irradiated	3	0	0	3
No information provided on the label regarding irradiation	70	1	2	67
Total	**76**	**1 (1.3)**	**2 (2.6)**	**73 (96.1)**

The most significant finding was that one sample which tested positive for irradiation by PPSL was not labelled appropriately, i.e. the sample was not in compliance with EU legislation. (Please note that labelling information was not provided for the second sample which tested positive for irradiation).

4.2.3 *Enterobacteriaceae* counts

In this study, *Enterobacteriaceae* counts were used as an indicator of irradiation or other similar treatments. For the purpose of this programme, the Commission proposed that samples with an *Enterobacteriaceae* count ≤100 cfu/g should be suspected of having been irradiated or submitted to similar treatments [35]..

In this study, *Enterobactericaeae* counts of ≤100 cfu/g were recorded for 73.9% (478/647) of single samples and 64% (16/25) of batch samples (Table 22) suggesting that these samples were irradiated or were submitted to similar treatments. (A breakdown of the *Enterobacteriaceae* results by health board is provided in Appendix 7). It is worth noting that this finding is significantly different to the results from the irradiation testing by PPSL suggesting that the samples were exposed to a treatment other than irradiation (e.g. heat treatment).

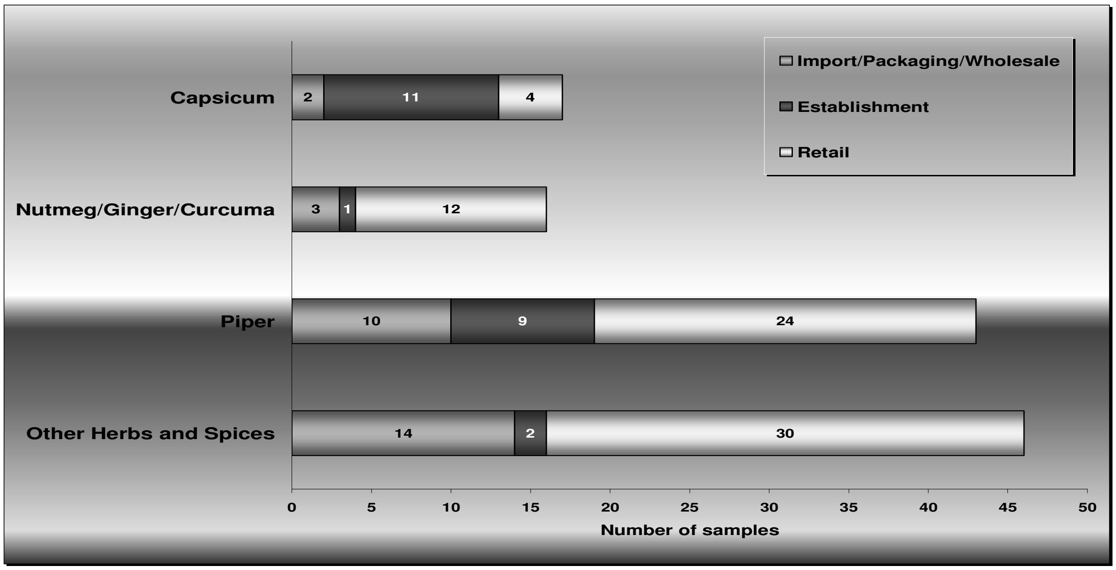
Table 22: *Enterobacteriaceae* results

		***Enterobacteriaceae* count**	
Batch/Single sample	**Total number**	**No. of samples with a count ≤100 cfu/g (%)**	**No. of samples with a count >100 cfu/g (%)**
Single	647	478 (73.9%)	169 (26.1%)
Batch	25	16 (64%)	9 (36%)

4.3 Aflatoxin Results

A total of 122 samples were taken at different stages of the distribution chain (Figure 3) and were analysed by the Public Analyst Laboratories.

Figure 3: Details of samples taken for aflatoxin testing



The results of the analyses for aflatoxins are summarised in Table 23, which shows the total numbers of samples having levels of aflatoxin B_1 below 2 µg/kg, between 2 – 5 µg/kg and above 5 µg/kg, together with the corresponding levels for total aflatoxins. Of the total 122 samples, only 2 samples (1.6%), one of ground nutmeg and one of chilli powder, were found to be non-compliant with the legal limit of 5µg/kg for aflatoxin B1, having levels of 5.6 µg/kg and 9.7 µg/kg aflatoxin B_1 respectively. The predominant aflatoxin detected in these samples was aflatoxin B1, and the legal limit for total aflatoxins was exceeded only in the chilli powder sample, with a level of 11.7 µg/kg aflatoxin B_1+B_2+G_1+G_2. Levels of aflatoxin were comparatively low in the remaining samples, with 15 of the 122 samples (12.3%) having levels of aflatoxin B_1 between 2 – 5 µg/kg and the remaining samples having levels below 2 µg/kg and frequently below the limit of analytical detection. Levels between 2 – 5 µg/kg occurred predominantly in those spices targeted in the EU Coordinated Programme for the Official Control of Foodstuffs 2004, namely pepper, chilli powder and nutmeg.

Table 23: Measured levels of aflatoxin B_1 and total aflatoxins in spices and herb samples

Sampling stage	Product identification	No. of samples	Level of aflatoxins (µg/kg)					
			B_1			Aflatoxin total		
			< 2	2 – 5	>5	<4	4 – 10	>10
Import of establishment for packaging or wholesaler	*Capsicum* spp.	2	2			2		
	Piper spp.	10	10			10		
	Nutmeg/ginger/curcuma	3	2	1		3		
	Other spices and herbs	14	13	1		14		
Establishment (using large amount of spices for food preparation)	*Capsicum* spp.	11	8	3		10	1	
	Piper spp.	9	9			9		
	Nutmeg/ginger/curcuma	1	1			1		
	Other spices and herbs	2	2			2		
Retail	Capsicum spp.	4	2	1	1	3		1
	Piper spp.	24	23	1		24		
	Nutmeg/ginger/curcuma	12	6	5	1	11	1	
	Other spices and herbs	30	27	3		28	2	

4.4 Heavy Metal Results

A total of 186 samples were analysed for heavy metal content (arsenic, cadmium, lead and mercury), comprising 107 of the samples analysed for aflatoxins plus an additional 79 samples. Although legal limits for these metals in spices and dried herbs have not been established, and it is therefore not possible to reach a definite conclusion about the safety of the samples taken relative to their content of these toxic metals, in general it can be stated that the levels were low. The majority of the results for mercury lay below the limit of detection (from 0.02 to 0.1 mg/kg). Levels of arsenic were in the range of less than 0.05 – 0.8 mg/kg, with only 7 of the 186 samples analysed having levels greater than 0.5 mg/kg. Results for cadmium were also low, in the range of less than 0.02 to 0.53 mg/kg. Somewhat higher levels of lead were detected, with 17 out of 186 samples analysed having levels greater than 1 mg/kg, and one sample of turmeric having a level of 21 mg/kg.

5. Conclusions

The findings of this study show that:

- The majority of samples were of a satisfactory microbiological status (94.1% of single samples and 84% of batch samples had an overall classification of satisfactory). However, pathogens (*Salmonella* spp. and *B. cereus*) were detected at unsatisfactory levels in a small number of samples.
- One sample which was irradiated was not labelled in accordance with EU legislation
- There was a high degree of compliance with the legal limits for aflatoxins
- Levels of the toxic metals arsenic, cadmium, lead and mercury were generally low.

The production of safe produce requires the implementation of food safety controls throughout the food chain. These include i) the implementation of good agricultural practices (GAP) during cultivation and ii) the implementation of good hygiene practices (GHP) at all stages throughout the food chain. The implementation of a food safety management system based on the principles of HACCP is an integral part of these practices.

As stated in the introduction, many herbs and spices are cultivated and harvested in countries with poor sanitary conditions and warm humid climates. These conditions increase the potential for microbial contamination and proliferation. Therefore it is imperative that importers implement strict supplier control. In addition, it is important that food handlers are aware of the microbiological risks associated with the use of herbs and spices as an ingredient in foodstuffs and implement the appropriate control measures (e.g. good temperature control during cooking, chilling and storage; and implementation of good handling and hygiene practices).

Recommended reading:

1) *Codex Code of Hygienic Practices for Spices and Dried Aromatic Plants* [2].
2) FSAI Code of Practice No. 4 '*Code of Practice for Food Safety in the Fresh produce Supply Chain in Ireland*' [36]

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7. APPENDICES
APPENDIX 1: Questionnaire

General Information:
- * EHO Name: ____________________
- * EHO Sample Reference Number (i.e. EHO's own personal reference number for the sample): ____________________

Sample Information:
- * Brand Name or Supplier Name and Address: ____________________
- * Type of Sample: ____________________
- * Category of sample
  - *Capsicum* spp. ☐
  - *Piper* spp. ☐
  - Nutmeg/ginger/curcuma: ☐
  - Other spices & herbs: ☐

Country of origin: ____________________

* Type of packaging: Loose ☐ Pre-packed ☐
* Did the public have access to the loose samples?: Yes ☐ No ☐
* Batch Number: ____________________ Not Available ☐
* Best Before date: ____________________ Not Available ☐
* Was the sample irradiated? (check label for info.): Yes ☐ No ☐ Not stated ☐

Premises Information
* EHO Premises Reference Number (i.e. EHO's own personal reference number for the premises): ____________________

* Type of premises

Import or Production or Packaging premises or Wholesaler	☐
Establishments using large amounts of herbs/spices for food preparation	☐
Retail	☐

Laboratory Reference Numbers:
Microbiological analysis

OFML Ref. No. for single sample: ____________________

OFML Ref. No's (x5) for batch sample: 1)__________ 2)__________ 3)__________ 4)__________ 5)__________

Chemical analysis:

Was the sample sent for chemical analysis: yes ☐ no ☐

If yes please state the PAL Ref. No.: ____________________

What tests were carried out on the sample?: aflatoxin ☐, heavy metals ☐, irradiation ☐

Bacteriological Safety

	Satisfactory	Acceptable	Unsatisfactory
Salmonella spp.	☐	N/A	☐
Bacillus cereus	☐	☐	☐
Clostridium perfringens	☐	☐	☐
Enterobacteriaceae	☐	☐	☐

N/A: Not Applicable

Enforcement Action *(please tick as many boxes as necessary):*
Reason for action: Bacteriological ☐ Chemical ☐ Both ☐

Type of action *(please tick as many boxes as necessary):*

None	☐		
Verbal warning	☐	Written warning	☐
Improved in house control required	☐	Product recall	☐
Administrative penalty	☐	Court action	☐
Other (Please specify)	____________________		

Appendix 2

Official Food Microbiology Laboratories (OFMLs)

OFML
Cherry Orchard Hospital
Mid-Western Regional Hospital
Public Analysts Laboratory, Dublin
Sligo General Hospital
St Finbarr's Hospital, Cork
University College Hospital, Galway
Waterford Regional Hospital

Public Analyst Laboratories (PALs)

PAL
Galway Public Analyst Laboratory
Cork Public Analyst Laboratory
Dublin Public Analyst Laboratory

Appendix 3

List of Health Boards

Health board	Abbreviation	No. of single samples submitted for analysis	No. of batch samples submitted for analysis
East-Coast Area Health Board	ECAHB	53 (8.2)	2 (8.0)
Midland Health Board	MHB	49 (7.6)	0 (0)
Mid-Western Health Board	MWHB	42 (6.5)	12 (48.0)
Northern Area Health Board	NAHB	57 (8.8)	0 (0)
North-Eastern Health Board	NEHB	41 (6.3)	0 (0)
North-Western Health Board	NWHB	63 (9.7)	0 (0)
South-Eastern Health Board	SEHB	121 (18.7)	3 (12.0)
Southern Health Board	SHB	42 (6.5)	8 (32.0)
South-Western Area Health Board	SWAHB	108 (16.7)	0 (0)
Western Health Board	WHB	71 (11.0)	0 (0)
		647 (100)	**25 (100)**

Appendix 4
Salmonella spp. results by health board

Health board	Single samples			Batch samples		
	Total no.	S	U	Total no.	S	U
ECAHB	53	53	0	2	2	0
MHB	49	49	0	0	0	0
MWHB	42	40	2	12	12	0
NAHB	57	57	0	0	0	0
NEHB	41	40	1	0	0	0
NWHB	63	63	0	0	0	0
SEHB	121	119	2	3	3	0
SHB	42	42	0	8	8	0
SWAHB	108	108	0	0	0	0
WHB	71	70	1	0	0	0
Grand Total	**647**	**641 (99.1%)**	**6 (0.9%)**	**25**	**25 (100%)**	**0 (0%)**

S: Satisfactory, U: Unsatisfactory

Appendix 5

B. cereus results by health board

Health board	Single samples				Batch samples			
	Total no.	S	A	U	Total no.	S	A	U
ECAHB	53	53	0	0	2	1	0	1
MHB	49	48	1	0	0	0	0	0
MWHB	42	31	11	0	12	10	2	0
NAHB	57	57	0	0	0	0	0	0
NEHB	41	40	0	1	0	0	0	0
NWHB	63	61	2	0	0	0	0	0
SEHB	121	116	5	0	3	3	0	0
SHB	42	40	2	0	8	7	1	0
SWAHB	108	106	1	1	0	0	0	0
WHB	71	69	2	0	0	0	0	0
Grand Total	**647**	**621 (96.0%)**	**24 (3.7%)**	**2 (0.3%)**	**25**	**21 (84.0%)**	**3 (12.0%)**	**1 (4.0%)**

S: Satisfactory, A: Acceptable, U: Unsatisfactory

Appendix 6

C. perfringens results by health board

Health board	Single samples				Batch samples			
	Total no.	S	A	U	Total no.	S	A	U
ECAHB	53	53	0	0	2	2	0	0
MHB	49	49	0	0	0	0	0	0
MWHB	42	42	0	0	12	12	0	0
NAHB	57	55	2	0	0	0	0	0
NEHB	41	40	1	0	0	0	0	0
NWHB	63	63	0	0	0	0	0	0
SEHB	121	116	5	0	3	3	0	0
SHB	42	41	1	0	8	8	0	0
SWAHB	108	108	0	0	0	0	0	0
WHB	71	71	0	0	0	0	0	0
Grand Total	**647**	**638 (98.6%)**	**9 (1.4%)**	**0 (0%)**	**25**	**25 (100%)**	**0 (0%)**	**0 (0%)**

S: Satisfactory, A: Acceptable, U: Unsatisfactory

Appendix 7

Enterobacteriaceae results by health board

Health board	No. of single samples			No. of batch samples		
	Total no.	≤100 cfu/g	>100 cfu/g	Total no.	≤100 cfu/g	>100 cfu/g
ECAHB	**53**	42	11	**2**	2	0
MHB	**49**	46	3	**0**	0	0
MWHB	**42**	18	24	**12**	8	4
NAHB	**57**	47	10	**0**	0	0
NEHB	**41**	33	8	**0**	0	0
NWHB	**63**	54	9	**0**	0	0
SEHB	**121**	83	38	**3**	2	1
SHB	**42**	34	8	**8**	4	4
SWAHB	**108**	78	30	**0**	0	0
WHB	**71**	43	28	**0**	0	0
Grand Total	**647**	478 (73.9%)	169 (26.1%)	**25**	16 (64%)	9 (36%)