

MONITORING & SURVEILLANCE SERIES



MICROBIOLOGY

Bacteriological and Chemical Safety of Ready-to-Eat Dried Seeds and Ready-to-Eat Nuts (10NS1)

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SUMMARY

This study investigated the microbiological, i.e. *Salmonella* spp. and *Escherichia coli*, and chemical, i.e. aflatoxin B1 and total aflatoxins, safety of 891 pre-packaged ready-to-eat (RTE) nut and seed products, on sale in the Republic of Ireland. Of the 891 samples analysed, 56.6% (504/891) were nut products, 42.6% (380/891) were seed products and 0.8% (7/891) were miscellaneous products.

The survey revealed that 0.3% (3/891) of samples were categorised 'unacceptable/potentially hazardous' for *Salmonella* spp., i.e. detected in 25g. Following serological examination of *Salmonella* spp. positive samples, *Salmonella* Agona was identified in one sesame seed sample and *Salmonella* Anatum in the other sesame seed sample. The products originated from India and Bolivia, respectively. *Salmonella* Marseille was identified in a melon (egusi) seed sample which originated from Africa. On investigation, the melon (egusi) seed sample was intended for use in cooking. However, as it was not labelled as requiring cooking, it was included in this survey. Cooking would be expected to kill *Salmonella*.

Escherichia coli was detected at 'unsatisfactory' levels, i.e. \geq 100 colony forming units/gram (cfu/g), in 0.3% (3/891) of samples. All 'unsatisfactory' samples were pre-packaged, edible, single seed products comprising two poppy seed samples and one pumpkin seed sample. All samples which were 'unsatisfactory' for *E. coli* had counts between 100 and 500 cfu/g. *E. coli* was also detected at 'acceptable' levels, i.e. 20 to <100 cfu/g in 0.8% (7/891) of samples.

All products with 'unsatisfactory' *E. coli* levels and 'unacceptable/potentially hazardous' levels of *Salmonella* were seed products. However, none of the laboratories detected both 'unacceptable/potentially hazardous' levels of *Salmonella* spp. and 'unsatisfactory' levels of *E. coli* in the same seed sample.

The findings of *Salmonella* and 'unacceptable' levels of *E. coli* in seeds highlight a requirement for guidance on good hygiene and manufacturing practices throughout all stages of RTE seed production but focussing particularly on the critical processes of cleaning and drying of seeds.

Two percent (18/891) of samples were analysed for aflatoxins B1 and total aflatoxins. All test results were categorised as 'satisfactory' for all of the mycotoxin test parameters. All 18 samples sent for chemical analysis were also found to have 'satisfactory' levels of *E. coli* and *Salmonella* spp.



ACKNOWLEDGEMENTS

The Food Safety Authority of Ireland (FSAI) wishes to thank the environmental health officers and the laboratory staff of the Health Service Executive (HSE) Official Food Microbiology Laboratories (OFMLs) and the Dublin Public Analysts Laboratory (PAL), Sir Patrick Duns, who participated in this survey.



ABBREVIATIONS

cfu	Colony Forming Unit
EU	European Union
EC	European Commission
EHO(s)	Environmental Health Officer(s)
EHS	Environmental Health Services
FSAI	Food Safety Authority of Ireland
g	Grams
HSE	Health Service Executive
ISO	International Standards Organisation
LIMS	Laboratory Information Management System
NSSLRL	National Salmonella, Shigella and Listeria Reference Laboratory
MS	Member State(s)
OFMLs	Official Food Microbiology Laboratories
PAL	Public Analyst Laboratory
RTE	Ready-to-Eat
RASFF	Rapid Alert System for Food and Feed
UK	United Kingdom
USA	United States of America



INTRODUCTION

Ready-to-eat (RTE) nuts and seeds are considered to be similar products in terms of their physical composition and production processes. Traditionally, RTE nuts and seeds have been a popular RTE food in Ireland, particularly due to their health benefits ¹⁻². RTE nuts and seeds are considered relatively safe due to their low moisture content despite little published data on their microbiological quality ¹. However, in recent years there have been a number of incidents worldwide relating to the detection of *Salmonella* spp. and other pathogens in RTE nuts and seeds ³⁻⁹. Other studies investigating the microbiological quality of RTE nuts, e.g. almonds, cashew, hazelnut, peanuts have also indicated the presence of *Salmonella* spp.¹⁰⁻¹⁴.

A 2007 United Kingdom (UK) study of retail edible dried seeds assessed their microbiological safety in relation to *Salmonella* contamination and levels of *Escherichia coli*, an indicator of faecal contamination ^{1,15}. Overall, *Salmonella* was detected in 0.6% (23/3735) of samples, of which over half were sesame seeds. Other seeds contaminated with *Salmonella* were linseed, sunflower, alfalfa, melon and mixed seeds. *E. coli* was detected in 9% (339/3735) of samples, with 1.5% (55/3375) containing 'unsatisfactory' levels ^{1,15}.

In Ireland, two food alerts were issued in 2009 leading to the recall of sesame seeds ¹⁶. In both alerts, products were recalled due to the detection of *Salmonella* spp. in seeds supplied via the UK to three retail chains in Ireland ¹⁶. There were no reports of illness. In 2010, two further food alerts were issued in Ireland, as a result of the current survey. These alerts led to recalls of sesame seed contaminated with *Salmonella* spp. ¹⁷ and *Salmonella* Anatum ¹⁸. Again no illnesses were reported.

Commission Regulation (EC) No 2073/2005 on microbiological criteria for foodstuffs ¹⁹, lays down food safety criteria for certain important foodborne bacteria, their toxins and metabolites, in specific foodstuffs. These criteria are applicable to products placed on the market during their entire shelf-life. In addition, the Regulation ¹⁹ sets down certain process hygiene criteria to indicate the correct functioning of the production process. While there are no specific microbiological criteria in the legislation for *Salmonella* spp. in RTE dried nuts and seeds; the presence of this pathogen in a RTE food would render it unsafe (as defined in Article 14 of Regulation 178/2002) ²⁰. Food business operators have a legal obligation to recall/withdraw unsafe food from the market (Article 19 of Regulation 178/2002) ²⁰.

In addition to bacterial contamination, growth of certain fungi (either in the field or post harvest) may lead to the production of mycotoxins in nuts and seeds. Mycotoxins are toxic to humans and animals. Maximum levels for mycotoxins in various foodstuffs have been established in European legislation, via Commission Regulation (EC) No 1881/2006 as amended ²¹. Regarding nuts, specific provisions are set for groundnuts, nuts and processed products intended for i) direct human consumption or ii) sorting or other physical treatments prior to human consumption (higher aflatoxin levels are permitted for products falling into the latter category). Regulation (EC) No 165/2010 ²² (amending Regulation 1881/2006) ²¹ sets limits for aflatoxins in oilseeds.



OBJECTIVES

- To examine the bacteriological safety (Salmonella spp. and E. coli) of RTE dried seeds and RTE nuts.
- To examine the chemical safety (Aflatoxins B1 and total aflatoxins) of RTE dried seeds and RTE nuts.



METHOD

This survey examined the bacteriological and chemical safety of RTE dried seeds and nuts available for sale in the Republic of Ireland, by testing for *Salmonella* spp., *Escherichia coli*, aflatoxin B1 and total aflatoxins.

Sample Collection

From April to July 2010 (inclusive) Environmental Health Officers (EHOs) collected samples as outlined in Table 1:

Table 1: Sam	ple Source.	Description	and Ana	alvsis ²³
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Sample Source ^{a-b}	Type of Analysis ^{c-f}
Establishments importing/wholesaling edible RTE dried seeds & nuts	Microbiological & chemical analysis
Retail establishments selling RTE dried seeds & RTE nuts, e.g. supermarkets, health food stores, farmers markets etc.	Microbiological analysis only

^a Samples collected included **Dried Seeds**, e.g. sesame, melon (egusi), poppy, pumpkin, linseed (flax), alfalfa, sunflower etc., including mixed dried seeds; **RTE Nuts**, e.g. almonds, brazil, cashew, hazel, macadamia, peanuts, pecans, pine nuts, pistachios, walnuts etc., including mixed nuts; **Loose** or **Pre-Packaged Samples**, **Shelled** or **Un-Shelled**

^b Samples collected excluded the following products containing seeds and/or nuts, e.g. health bars, cereals, peanut butter etc; Mixes of seeds and/or nuts containing dried fruit or any other ingredient; Coated seeds and/or nuts, e.g. coated in yoghurt, chocolate, carob, chilli etc; Salted and/or seasoned seeds and/or nuts, e.g. salted and dry roasted ²³

 $^{\circ}$ Samples for microbiological analysis were a minimum of 75g

^d Samples for microbiological and chemical analysis were obtained from the same production batch

^e For chemical analysis, given the potentially heterogeneous nature of mycotoxin contamination within a given batch of a commodity, sampling was undertaken in such a way as to ensure it was fully representative. Commission Regulation 401/2006 ²⁴ sets out the requirements for sampling and analysis of products in respect of the controls of mycotoxins established in European legislation, Commission Regulation 1881/2006 as amended ²¹. The provisions of Regulation 401/2006 (European Commission) are implemented in Ireland via S.I. 412 of 2006 ²⁵. The FSAI guidance on the sampling of products for the presence of mycotoxins ²⁶ was followed during sampling for this survey.

^f For chemical analysis, samples were obtained by EHOs normally involved in sampling for mycotoxins under the National Chemical Sampling Plan.



Sample Number

As for previous national microbiological surveys, arrangements regarding the number of samples for microbiological analysis were made locally between the Environmental Health Services (EHS) and the Health Service Executive (HSE) Official Food Microbiology Laboratories (OFMLs).

Sampling Procedure

EHOs took a single sample of each variety of seeds and nuts from each available brand. For a small number of products, representative samples were taken in line with the requirements of Commission Regulation (EC) No 401/2006 for aflatoxin analysis ²⁴. If problems were encountered obtaining adequate sample numbers, more than one sample of each variety was taken, provided the samples were from different production batches. Following collection, all samples were stored and transported to the laboratory under appropriate conditions.

EHOs were requested to complete a questionnaire for each sample collected (Appendix 1). Where samples from the same production batch were submitted for both microbiological and chemical analysis, only one questionnaire was completed. In the case of repeat samples, analysis as well as questionnaires weren't included to ensure the sample was not included in the survey.

Sample Analysis

Microbiological Analysis

Microbiological analysis was undertaken at the seven HSE-OFMLs. Samples were prepared for analysis using International Standards Organisation (ISO) 6887-4:2003²⁷. Samples were then analysed for *Salmonella* spp. (detection) using ISO 6579:2002²⁸ and *E. coli* (enumeration) using ISO 16649-2:2001²⁹. If a sample tested positive for *Salmonella* spp., the isolate was sent to the National Salmonella, Shigella and Listeria Reference Laboratory (NSSLRL), Galway for confirmation of speciation.

Chemical Analysis

Sample preparation and analysis for the presence of mycotoxins was undertaken by the Dublin PAL, for aflatoxin B1 and total aflatoxins.

Data Collection

Data were submitted to the FSAI where they were collated and quality assured. The laboratory report data were submitted via Laboratory Information Management System (LIMS) extracts and the questionnaire data were received in hard copy format.

Interpretation and Classification of Results

Microbiological Results

There are no *Salmonella* or *E. coli* standards specified in legislation for nuts and seeds ¹⁹. Therefore, the results are assessed against the following RTE microbiological guidelines given in Table 2 ³⁰:



Table 2: Guidelines for the Microbiological Quality of RTE Nuts & Seeds					
Guideline	Satisfactory	Acceptable	Unsatisfactory	Unacceptable/ Potentially Hazardous	
Salmonella spp.	Not detected in 25g	N/A	N/A	Detected in 25g	
E. coli	< 20 cfu/g	20 to < 100 cfu/g	≥ 100 cfu/g	N/A	

Table 2: Guidelines for the Microbiological Quality of RTE Nuts & Seeds

Chemical Results

The chemical results for aflatoxin analysis in nuts were assessed against the maximum levels set out in Commission Regulation (EC) No 1881/2006²¹ as amended, to ensure they complied with the legislative limits. Results for mycotoxin analysis in seeds were assessed against the maximum levels set out in Commission Regulation (EC) No 165/2010²². These limits are also available in the FSAI report "Legislation on Chemical Contaminants" ²⁶.

Statistical Analysis

Descriptive and statistical analysis of the data was undertaken using Microsoft Excel and SPSS version 14.0. Relative proportions were compared using Chi Square (χ^2) and Fisher's Exact Test. A probability value of less than 5% i.e. p ≤ 0.05 level was defined as significant.



RESULTS AND DISCUSSION

Summary Data

A total of 891 samples were analysed at the OFMLs. Of the 891 samples analysed, 56.6% (504/891) were nut products, 42.6% (380/891) were seed products and 0.8% (7/891) were miscellaneous products.

Samples included 17 different varieties of seed and 10 varieties of nut. The most commonly collected varieties were sunflower seeds 12.2% (109/891), Brazil nuts 9.1% (81/891) and pumpkin seeds with 8.9% (79/891) of total samples. The most commonly sampled single variety nut and seed products are outlined in Table 3 and represent 74.5% (664/891) of all samples analysed during the survey.

Nut or Seed Variety	Number of Samples	Percentage of Total
Sunflower Seed	109	12.2
Brazil Nuts	80	9.0
Pumpkin Seed	79	8.9
Sesame Seeds	76	8.5
Hazelnuts	69	7.7
Cashew Nuts	61	6.8
Almonds	60	6.7
Walnuts	53	5.9
Peanuts	39	4.4
Pine Nuts	38	4.3
All Others	227	25.5
Total	891	100%

Table 3: Most Commonly Sampled Single Variety Nut and Seed Products (n=891)

The majority of samples, approximately 71%, taken during the survey were from retailers, with 41.5% (370/891) and 30.2% (269/891) for nuts and seeds, respectively. Minor quantities of samples were taken from distributors and transporters 2.5% (22/891), manufacturers and packers 2% (18/891) and the service sector 1.7% (15/891). The food chain stage was not indicated by EHOs in 22.1% (197/891) of both seed and nut samples taken during the survey.

Microbiological Data

Salmonella Contamination

Overall, 99.7% of all RTE dried seed and nut samples were 'satisfactory' (Table 2) for *Salmonella* spp. *Salmonella* spp. was only detected in 0.3% (3/891) of samples surveyed. The three samples positive for *Salmonella* spp. in 25g, i.e. 'unacceptable/potentially hazardous' were all pre-packaged, edible, single seed products, i.e. 2 x sesame seeds and 1 x melon (egusi) seeds. Following serological examination, *Salmonella* Agona was identified in one sesame seed sample and *Salmonella* Anatum in the other sesame seed sample, with the products originated from India and Bolivia, respectively. *Salmonella* Marseille was identified in the melon (egusi) seed sample which originated from Africa. In African cuisine, melon (egusi) seeds, are typically used in cooking, however because the product was not labelled as requiring cooking it was included in this survey. Cooking would be expected to kill *Salmonella*. Table 4 provides a summary of information related to the three samples were *Salmonella* spp. was detected and the actions taken thereafter.



General Information	Sample 1	Sample 2	Sample 3
Lab Performing Analysis	Cherry Orchard (OFML)	Sir Patrick Duns (PAL)	Waterford (OFML)
Sample Description	Sesame seeds	Melon (egusi) seeds ^a	Sesame seeds
Species Detected	Salmonella Agona	Salmonella Marseille	Salmonella Anatum
Food Chain Stage	Retail	Retail	Retail
Country of Origin	India	Africa	Bolivia
Type of Packaging	Pre-packaged	Pre-packaged	Pre-packaged
Actions Taken ^b	Contaminated seeds were recalled and food alerts issued as well as an EU alert via the Rapid Alert System for Food and Feed (RASFF).	 Repeat Sample - 'satisfactory' Salmonella Result Remaining products were removed from the market and re-labelled with cooking instructions. 	Contaminated seeds were recalled & food alerts issued as well as an EU alert via the RASFF.

Table 4: Summary of Information Related to Salmonella spp. Positive Samples

^a Typically sold in African stores and consumed cooked in African dishes, however because the product was not labelled as requiring cooking it was included in this survey ^b See Appendix 2 for further details

Escherichia coli Contamination

Escherichia coli was included in the current survey as it is considered a useful indicator of faecal contamination of food products, and as such indicates the potential risk of contamination with faecal pathogens ^{19, 31}. Overall, 99.7% of all RTE dried seed and nut samples were 'satisfactory' or 'acceptable' (Table 2) for E. coli. E. coli was only detected at 'unsatisfactory' levels in 0.3% (n=3/891) of samples surveyed (Table 5).



	-	
Result Categorisation	Number of Samples	Percentage of Total Samples
Unsatisfactory ^a	3	0.3%
Acceptable ^b	7	0.8%
Satisfactory $^{\circ}$	881	98.9%
Totals	891	100%

Table 5: Summary of E. coli Results (n=891) Image: Coli Results (n=891)

^a Limit of ≥ 100 cfu/g

^b Limit of 20 to < 100 cfu/g

 $^{\circ}$ Limit of < 20 cfu/g

The majority of samples 98.9% (881/891) were 'satisfactory' for levels of *E. coli* (Table 5). All three 'unsatisfactory' samples outlined in Table 5 were pre-packaged, edible, single variety seed products and had *E. coli* counts between 100 and 500 cfu/g. Table 6 provides a summary of information related to the three samples where 'unsatisfactory' levels of *E. coli* were detected and the follow-up action that was taken on foot of the finding.



General Information	Sample 1	Sample 2	Sample 3
Lab Performing Analysis	Limerick (OFML)	Limerick (OFML)	Waterford (OFML)
Sample Description	Pumpkin seeds	Poppy seeds	Poppy seeds (organic)
Level Detected	100 cfu/g	500 cfu/g	260 cfu/g
Food Chain Stage	Retail	Retail	Retail
Country of Origin	China	More than one country	More than one country
Type of Packaging	Pre-packaged	Pre-packaged	Pre-packaged
Actions Taken ^a	 Repeat Sample (Lab Ref F- 0562/10) – 'satisfactory' Notified manufacturer. 	 Incident was reported to rapid alert. As there is no legal criterion for <i>E. coli</i> in seeds, no withdrawal or recall was required in this case. However, further Investigation was carried out at the food business operator by local EHOs. 	Notified the FSAI, the wholesaler & the manufacturing company.

Table 6: Summary of Information Related to Unsatisfactory levels of E. coli

^a See Appendix 2 for further details

Escherichia coli were also detected at 'acceptable' levels in 0.8% (7/891) of samples (Table 5). The *E. coli* counts for these 7 products ranged from 20 to 70 cfu/g. All 7 products were pre-packaged and comprised 5 seed variety products, i.e. 3 x pumpkin seeds, 1 x poppy seeds and 1 x linseed and 2 nut variety products, i.e. 1 x pine nuts and 1 x cashew nuts having 'acceptable' levels of *E. coli*.

All products with 'unsatisfactory' *E. coli* levels and 'unacceptable/potentially hazardous' levels of *Salmonella* were seed products. None of the samples from the current 10NS1 survey, analysed by the laboratories, detected both 'unacceptable/potentially hazardous' levels of *Salmonella* spp. and 'unsatisfactory' levels of *E. coli* in the same seed sample.



Chemical Data

Two percent (18/891) of samples from the current survey were sent by EHOs for chemical analysis of total aflatoxins and aflatoxin B1. None of the samples analysed in the survey for aflatoxin contamination exceeded the maximum levels for these contaminants set out in Commission Regulation (EC) No 1881/2006²¹, so no additional or confirmatory analyses were required. As such, all test results were categorised as 'satisfactory' for all of the mycotoxin test parameters. All 18 of the samples sent for chemical analysis were also found to have 'satisfactory' levels of *E. coli* and *Salmonella* spp.

Questionnaire Data

EHOs were required to complete a questionnaire at the time of sampling which provided additional information on the samples taken in the survey. The following findings relate to questionnaires for samples analysed at all laboratories involved in the survey outlined in Table 1.

Sample Type

A total of 823 questionnaires were received in respect of the 891 samples analysed, a response rate of 92%. A breakdown of sample types collected based on the 823 questionnaires received indicated that 54.2% (446/823) of samples were nuts, 40.3% (332/823) seeds, 3.4% (28/823) mixed nuts and 2.1% (17/823) mixed seeds.

The most commonly sampled nuts were Brazil nuts making up 9.2% (76/823) of all samples for which a questionnaire was received. This was followed by hazelnuts, almonds and cashew nuts making up 7.5% (62/823), 7.0% (58/823) and 6.8% (56/823) respectively of all samples for which a questionnaire was received.

The most commonly sampled seeds were sunflower seeds making up 11.8% (97/823) of all samples for which a questionnaire was received. This was followed by sesame seeds, pumpkin seeds and linseed (flax) seeds making up 8.7% (72/823), 8.5% (70/823) and 5.1% (42/823) respectively of all samples for which a questionnaire was received.

The figures for the most commonly sampled nuts and seeds for which a questionnaire was received, are broadly in line with results obtained from laboratory reports outlined in Table 3.



Country of Origin

Samples taken during the current survey originated from at least 32 countries. Table 7 provides a breakdown of the countries of origin, when provided, for which a questionnaire was received.

Country of Origin	Number of Samples	Percentage of Total Questionnaires Received
China ^ª	116	14.1
United States of America (USA) ^a	72	8.7
Turkey ^a	40	4.9
Non European Union Countries (excluding China, USA & Turkey)	161	19.6
European Union (excluding Ireland)	90	10.9
Product of more than one country	50	6.1
Ireland ^b	29	3.5
Not Stated	265	32.2
TOTAL	823	100%

Table 7: Breakdown of Country of Origin (when provided) (n=823)

^a The most common countries of origin from the survey based on questionnaires returned.

^b While 3.5% of samples had their country of origin recorded as Ireland, many of these nut and seed products are not grown or harvested in Ireland, e.g. pistachios, peanuts and almonds. This was also the case for some products originating in the United Kingdom. ^c There was no significant relationship (p>0.05) between country of origin and 'unacceptable/potentially hazardous' levels of *Salmonella spp.* and 'unsatisfactory' levels of *E. coli.*

Packaging & Labelling

Of the 823 questionnaires received, 95.1% (783/823) of products surveyed were pre-packaged, 1.2% (10/823) were loose but in 3.6% (30/823) of cases the packaging status was unstated. In the case of products which were pre-packaged, 5.7% (47/823) were packaged in the establishment where they were sampled. Brand and/or manufacture information was provided on the majority of samples 97.8% (805/823) with only 2.2% (18/823) not providing this information. 95.6% (787/823) displayed a 'best-before' date and 86.0% (708/823) displayed a production batch number for traceability purposes.



CONCLUSIONS

Results from this survey demonstrated that the vast majority 99.3% (885/891) of RTE dried nuts and seeds were of 'satisfactory' or 'acceptable' microbiological quality. However, a small proportion 0.7% (6/891) of samples did not meet the guidelines for microbiological quality of RTE nuts and seeds set out in Table 2. Three of these samples that were contaminated with *Salmonella* spp. were categorised as 'unacceptable/potentially hazardous'. Any RTE foods contaminated with *Salmonella* spp. are considered unsafe and of unacceptable risk because they do not receive any further cooking by the consumer that would kill the *Salmonella* ²⁰. Where *Salmonella* spp. was detected and/or 'unsatisfactory' levels of *E. coli* from samples examined as part of this survey, the appropriate action was taken to protect public health. Contaminated seeds (Table 4) were recalled and food alerts issued as well as an EU alert via the Rapid Alert System for Food and Feed (RASFF). None of the samples analysed in the survey for aflatoxin contamination exceeded the maximum levels for these contaminants set out in Commission Regulation (EC) No 1881/2006 ²¹.

The contaminated products identified in this survey were all pre-packaged and sampled from different retailers (Tables 4 & 6). This may imply that the results are not simply related to poor hygiene and/or manufacturing practices by a single producer, but rather a more widespread issue with hygiene and manufacturing practices in the industry and the different countries supplying the products.

The findings of this current survey have highlighted the potential public health risks associated with the consumption of RTE seeds due to contamination with *E. coli*, but more importantly *Salmonella* spp. There is a requirement for guidance on good hygiene and manufacturing practices throughout all stages of RTE nut and seed production. However, there is little published guidance on good hygiene practices for use by the RTE seed producing industry. From literature, it appears that critical control points for controlling the microbiological quality of RTE seeds are the cleaning and drying processes used in production ^{1,32,33}. The Grocery Manufacturers Association in the United States has published an industry handbook for the safe processing of nuts ³⁴. In addition, good hygiene practice is necessary when these products are being re-packaged into smaller packets for retail sale.



APPENDIX 1: QUESTIONNAIRE 10NS1

<u>1. General Information</u> : EHO Name:					
2. EHO Reference Number & Laboratory Reference Number:					
Please complete appropriate row: Sample submitted Please tick		EHO & Laboratory	Reference number		
for the following analysis	appropriate box	Microbiological analysis	Chemical analysis		
Microbiological <u>and</u> chemical analysis		EHO Ref No.:	EHO Ref No.:		
Microbiological analysis <u>only</u>		EHO Ref No.:	N/A		
	Central Distribut	col): tion Centre □; Retail establishr I, Health store □, Farmers market □			
3. Sample description (Se Sample description)	_ ee section 4 of P Seeds	rotocol): xed Seeds	uts 🗆		
(please specify	□, Poppy □,	ties: Pumpkin □, Linseed (Flax) □, Alfa or No □ or Don't know □	alfa 🔲, Sunflower 🗍, Other 🗍,		
If nuts, please specify the	e variety/varieti ashew □, Haze	es: ⊵l □, Macadamia □, Peanuts □, F	Pecans \Box , Pine nuts \Box , Pistachios		
Were the nut shells inclu	ded?: Yes 🗌 o	or No 🗌 or Don't know 🗌			
4. Other sample information: or Not Stated Country of origin:					
5. Follow-up Action (See Action required based on: r		otocol): esults	both \Box or no action required \Box		
Where follow-up action was required please provide details of the action taken: Product recall ; Product withdrawal ; Repeat sample (Please provide Lab. ref. no.: no.:) Other action (please specify:))					

Please note: 1) EHOs must complete this questionnaire for all samples, 2) Sections 1 to 4 should be completed by the EHO at the time of sampling 3) All questions are mandatory 4) All questionnaires must be returned to the FSAI by 10th Sept. 2010



APPENDIX 2: RECOMMENDED FOLLOW-UP ACTION BASED ON MICROBIOLOGICAL RESULTS

	Action to be taken
Salmonella species Detected in 25g	 The following must be notified immediately of the result: The food business operator from the establishment where the nuts or seeds were sampled (where this establishment is part of a retail multiple, the relevant person in headquarters should also be notified). The producer / packer (i.e. if different from above). The official agency responsible for the production / packaging. If this is the HSE, please contact the PEHO in this area. If the nuts / seeds are produced / packaged outside of the Republic of Ireland please contact FSAI. The FSAI (notification through the rapid alert system: rapidalert@fsai.ie). If the product is still on the market (based on its date of minimum durability), the FBO must recall the product or batch of foodstuff in accordance with Article 19 of Commission Regulation (EC) No 178/2002. See FSAI GN 10 Product Recall and Traceability for guidance on product withdrawal and recall. (Note: Revised version due for publication February 2010). The FBO must immediately investigate the cause of the unsatisfactory result and take the necessary corrective action to prevent re-occurrence. Furthermore, the food safety management system must be reviewed and amended if necessary (this may include amendments to microbiological sampling and testing). Note: The possible cause of the unsatisfactory result may have occurred due to inadequate processing or cross contamination. This may have happened at one or more points in the food chain - at retail level particularly if the nuts / seeds are being handled or sold loose or further back in the food chain at the point of processing / packing or the nuts and seeds may have been contaminated at harvesting. The nature and extent of the action required by the individual food business operators in the food chain will need to be considered on a case by case basis.
<i>Escherichia coli</i> ≥ 100 cfu/g	 The following must be notified of the result in the first instance: The food business operator from the establishment where the nuts or seeds was sampled (where this establishment is part of a retail multiple, the relevant person in headquarters should also be notified). The producer / packer (i.e. if different from above). These results indicate possible poor hygiene practices and must be investigated. The FBO must investigate the cause of this result and take the necessary corrective action to prevent re-occurrence. Furthermore, the food safety management system must be reviewed and amended if necessary (this may include amendments to microbiological sampling and testing). Note: The possible cause of this result may be due to poor handling, cross contamination etc. This may occur at retail level particularly if the nuts / seeds are being handled or sold loose or further back in the food chain. The nature and extent of the action required by the individual FBOs in the food chain will need to be considered on a case by case basis. Where follow-up sampling is undertaken for the purposes of enforcement action, those samples must be taken formally in accordance with legislative requirements (S.I. No. 747 of 2007).



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Abbey Court, Lower Abbey Street, Dublin 1.

Advice Line: 1890 336677 Telephone: +353 1 817 1300 Facsimile: +353 1 817 1301 Email: info@fsai.ie Website: www.fsai.ie