



## **Survey of Tortilla Chips and Taco Shells for Genetically Modified Ingredients**

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## **1. SUMMARY**

The Food Safety Authority of Ireland (FSAI) has completed a survey to determine the levels of genetically modified (GM) maize ingredients in tortilla chips and taco shells on sale in Ireland. The results confirm the findings of an earlier preliminary survey that detected GM ingredients in a range of different foods. This is the first in a series of surveys planned by the FSAI to determine the level of GM contents in different foods and to ensure that labelling regulations are adhered to.

This study focused on taco shells and tortilla chips due to concerns over reports of unlicensed GM ingredients in similar foods on the international market. The results show that GM maize ingredients were present in 19 of the 26 samples tested. The majority had levels below 0.1% which is considerably less than the 1% threshold level that triggers the labelling requirement. The specific variety of GM maize present in some samples was determined, but this was not possible in all cases due to limitations of the analytical methods.

There are no known health implications associated with the presence of such GM ingredients in these products. Because none of the samples contained GM maize at or above the threshold limit of 1%, specific GM labelling was not required on these foods. Furthermore, those that were individually identified are all maize varieties that have been approved for food use within the EU and, therefore, have already undergone rigorous food safety assessment.

## **2. PURPOSE OF THIS STUDY**

The FSAI's mission is to protect consumer health by ensuring that food consumed, distributed, marketed or produced in the State meets the highest standards of food safety and hygiene. In order to protect consumers and provide information with which they can make informed choices, the FSAI carries out routine surveillance to monitor compliance with food regulations.

On January 1<sup>st</sup> 2001, the FSAI became the competent authority for novel food, including GM food, in Ireland. As such, the FSAI is responsible for the implementation of EC Regulations concerning the marketing and labelling of GM foods.

The FSAI has embarked on a campaign to monitor food on sale in Ireland for GM ingredients and to ensure that appropriate labelling is being applied. This initial study focused on tortilla chips and taco shells in the light of claims made in the UK and US concerning unauthorised maize ingredients detected in similar foods.

Further focused studies are planned and, to that end, the FSAI is obtaining samples of other food groups containing soya and maize ingredients for similar analysis.

### **3. BACKGROUND**

In September 2000, an article published in the *Washington Post*, US reported that StarLink™ corn had been found in Taco Shells in the US. This type of GM corn, marketed by Aventis, was approved by the US Environmental Protection Agency (EPA) for use in animal feed in 1998, but not for human consumption. The feed approval was granted on condition that a stewardship scheme was put in place to ensure that the corn variety was only grown and used for animal feed purposes. Approval for food use was not granted because of unresolved concerns that the protein produced as a result of the genetic modification (Cry9c) might be allergenic. Although no evidence of allergenicity came from animal studies, it was concluded that there was insufficient information on the allergenic potential of the GM protein to license it for human consumption.

However, consumer groups claimed that independent testing had detected the unapproved corn in taco shells sold in the US and they asked the Food and Drug Administration (FDA) to recall the products in question. Following a full investigation the FDA confirmed that the Cry9c protein was detectable in taco shells supplied to them by the manufacturer, Kraft Foods and the products were withdrawn from sale. A few weeks later, US-based Safeway announced the voluntary withdrawal of its own brand of taco shells for the same reason. Since then, Aventis has withdrawn registration of StarLink™, which means that it can no longer be commercially cultivated or traded within the US. The US Department of Agriculture (USDA) is in the process of purchasing any remaining StarLink™ stocks, which will either be destroyed or used in feed or industrial processes. The US EPA is allowing a limited time period for this variety to be diluted out of existing food, feed and seed stocks.

In November 2000, tests commissioned by Friends of the Earth (FoE), UK detected two types of unapproved GM maize in four supermarket own-brand tortilla chips. GA21 Roundup Ready maize was found in Safeway and Asda brands, and traces of another type of GM maize (DBT418) were found in own-brand products sold by Sainsbury's and Tesco. Both varieties of maize are manufactured by Monsanto and are licensed for use in the US, but are not yet approved for sale within the EU. The UK Food Standards Agency investigated the claims made by FoE and independent analysis failed to detect unauthorised GM ingredients in any of the products.

#### **3.1 Maize**

Maize (corn) is one of the world's biggest crops, with a yearly harvest in excess of 560 million tonnes. The majority of maize is used in animal feed, with the remainder used for human food. One variety, a sweet corn, is eaten as a vegetable, while two other varieties, hard (flint) and soft (dent) maize are used in food processing. Hard maize is grown in South America and is mainly used in foods such as bakery products and breakfast cereals. Popcorn is a specific variety of this type of maize grown mainly in the US. Much of the soft maize is grown in Europe and is usually processed into flour, oil, starches and glucose syrups, which are used in many foods including snack foods, margarine and salad dressings.

Maize, like other crops, is susceptible to injury from pests such as the corn borer, which can result in significant yield loss. New varieties of maize have been genetically modified to express resistance to this pest and to select herbicides, thereby negating the need for topical pesticide applications. While these varieties are common in the US and Canada, the EU limits cultivation to a few varieties, and only ingredients derived from the maize varieties listed in Table 1 have been approved for food use within the EU.

**Table 1: Maize Varieties Approved for Food-Use in the EU**

<b>Company</b>	<b>GM Maize Variety</b>
Aventis	T25
Monsanto	MON810
Novartis	CG00526-176, Bt11
Pioneer	MON809

### **3.2 Labelling Threshold**

According to new EC legislation<sup>1</sup>, the labelling of food or food ingredients produced from, or containing licensed genetically modified organisms (GMOs) must indicate the inclusion of these ingredients where they are present at or above a level of 1%. This threshold level came into effect on 10 April 2000 and applies at the level of each ingredient, not the whole food. This regulation applies only to food ingredients proven to be from non-GM sources, where any GM content is as a result of adventitious contamination. The labelling threshold does not apply to GM containing food ingredients of unknown origin and such ingredients require labelling regardless of the level of GM content. Food containing additives or flavourings derived from, or containing GMOs must also be labelled accordingly under similar new EC legislation.<sup>2</sup>

### **3.3 QUID**

In certain circumstances it is necessary to state on food labels the percentage quantity of an ingredient used in the manufacture/preparation of the foodstuff. One example is where the ingredient is included in the name of the food, e.g. 'Corn Chips'. In this case it is necessary to state the quantity (%) of corn/maize present on the label. There are a number of other circumstances where such quantitative ingredient declaration (QUID) labelling is required, as well as those where it is unnecessary. Further information on this issue is provided in an EC document<sup>3</sup>. The Irish Business and Employers Confederation (IBEC) has also produced guidance for manufacturers and suppliers on the introduction of QUID in Ireland.

### **3.4 Monitoring Foods on the Irish Market**

The FSAI carried out a pilot survey at the end of 1999 to determine the GM status of processed foods containing soya and maize ingredients. This was not a comprehensive survey and did not include all brands or retail outlets. Products consisting of, or containing soya or maize, or their derivatives were randomly selected from the shelves. Given the preliminary and incomplete nature of this survey, the results (Appendix A) were not published, but rather

used as a reference for a future national surveillance campaign. Advances in analytical technology have led to the development of more sensitive and reliable protocols for the analysis of a wide range of GM food ingredients enabling comprehensive studies like that reported here on taco shells and tortilla chips.

### **3.5 Current Study**

This was a comprehensive survey of tortilla chips and taco shells on sale in Ireland. Samples of each of the 15 brands of these products known to be on the Irish market were obtained either directly from the relevant retailers or suppliers, or purchased 'off the shelf' by the FSAI. All samples were in pre-packaged form, as would be available to the consumer, but were sent 'blind' (FSAI identification markings only) for analysis. Given that these were processed foods, a multiple sampling regime was not necessary since such food can be considered to contain a homogenous mix. However, to eliminate any possibility of bias, two to three packets (depending on size/weight) from different lots/batches of each variety of the different brands were sampled where possible.

Due to the number of flavours included in some brands, many of the 26 samples sent for analysis comprised equal amounts of 2-3 flavours. Analysis involved a general screen for GM DNA, followed by more specific testing of positive samples to identify, where possible, the specific variety and relative amount of GM maize present.

## **4. METHODOLOGY**

Samples were analysed for GM material using a DNA based detection method, the Polymerase Chain Reaction (PCR). This system allows the detection and amplification of specific segments of DNA and can be used to characterise GM material in food. All samples, composite or otherwise, were homogenised to ensure that the sub-samples taken for analysis were representative of the entire sample.

DNA was then isolated from the samples and tested for the presence of unmodified maize DNA to determine the integrity of the extracted DNA. Where unmodified maize DNA was detected, samples were further screened for the presence of GM DNA. Upon detection of GM DNA further tests to identify specific genes used in the different varieties of GM maize were carried out. With this system, all samples were screened by at least 3 PCR tests, each of which had to confirm the others before a final result was reported.

All samples were extracted in duplicate and each PCR carried out twice to ensure reproducibility of results. A series of controls were routinely used to ensure that the extraction and PCR protocols were adequate and reliable. The positive control was a certified GMO standard obtained from the Institute for Reference Materials, Belgium and was used to monitor the efficiency of DNA extraction and PCR. Negative controls were used to ensure the absence of contamination and to avoid false positive results. These qualitative tests for GMO ingredients in processed foods can typically detect below 0.01% GM DNA

in a sample (as determined by international reference materials), but the sensitivity of the methods may vary between samples. The methods used have been validated by the Joint Research Centre of the EU.<sup>4</sup>

Where sufficient GM DNA was present in a sample, quantitative analysis was undertaken to determine the relative content of GM maize DNA. This was performed using Real-Time PCR (Roche Lightcycler) employing a fluorescence based (Scorpion™) detection system.<sup>5</sup>

For a complete description of the methods, see Appendix B.

## **5. RESULTS**

The data presented in Table 2 represents the GM status of each sample and, where possible, the variety and relative quantity of GM maize DNA present. The results provided relate solely to the sample tested and may not necessarily be representative of the general GM status of that product.

### **5.1 GM Content**

The presence or absence of GM maize DNA in each of the samples analysed is shown in Table 2. Positive (+) indicates that GM DNA was present in the sample and negative (-) that GM DNA was not detected. Trace (Tr) indicates the presence of trace amounts of GM DNA, i.e. a very weak signal was observed for some, but not all PCR reactions. It should be noted that, although some of the signals for the samples marked '+' were quite weak, positive signals were detected for each of the three PCR tests performed, thus confirming the presence of GM DNA in the sample.

### **5.2 GM Variety**

GM variety testing was performed on those samples in which a positive, or trace GM signal was detected. This PCR test identifies specific genes used in GM maize crops. While the identity of the specific GM maize variety was determined for a few samples (Table 2), the majority were shown to belong to a group (or category) of varieties, as described after Table 2. Of the varieties listed in these categories, all but two (MON801 and MON802) are approved for food use within the EU. Samples were not tested for the presence of the licensed GM maize variety T25 due to technical limitations.

### **5.3 Percentage of GM DNA**

Positive samples were also subjected to quantitative analysis to determine the amount of GM maize DNA present. These results (Table 2) represent the percentage of GM maize DNA relative to non-GM maize DNA in each sample tested. However, due to the limitations of the technology with respect to low levels of target DNA, it was not possible to quantify the amount of GM maize DNA in trace positive samples, so the percentage is simply recorded as trace (Tr).

**Table 2: Results of Tortilla Chip and Taco Shell Survey**

<b>Brand</b>	<b>Product</b>	<b>GM content<sup>a</sup></b>	<b>GM variety<sup>b</sup></b>	<b>GM %</b>
Adelita	Tortilla chips - natural - BBQ	-	NA	NA
Adelita	Tortilla chips - chili - nacho cheese	-	NA	NA
Amaizin'	Bio corn chips - natural - sesam	-	NA	NA
Amaizin'	Bio corn chips - chili - paprika	-	NA	NA
Casa Fiesta	Authentic tortilla chips Cheese flavour tortilla chips Chili flavour tortilla chips	Tr	Bt176	Tr
Casa Fiesta	Taco shells	+	Category A	<0.1
Discovery	Traditional taco shells	Tr	NI	Tr
Doritos	Dippas - dipping chips - lightly salted - hint of lime	Tr	Category B	Tr
Doritos	3Ds corn snacks - tangy cheese - cool spice - texas paprika	Tr	Category B	Tr
Doritos	Corn chips - cool original - tangy cheese	Tr	NI	Tr
Evernat	Natural tortilla chips Chilli tortilla chips	-	NA	NA
Old El Paso	Taco shells (dinner kit)	+	Category A	<0.1
Old El Paso	Nachips	+	Category A	<0.1
Old El Paso	Taco shells	+	Bt176	<0.1
Phileas Fogg	Authentic tortilla tostaditas - chilli - thankfully cool	+	NI	<0.1
Phileas Fogg	Authentic tortilla tostaditas - nacho cheese - devilishly hot	Tr	NI	Tr
Phileas Fogg	Authentic tortilla chips	+	NI	0.5
Poco Loco	Natural tortilla chips Nacho cheese tortilla chips	-	NA	NA
Poco Loco	Chili tortilla chips Barbecue tortilla chips	-	NA	NA



Brand	Product	GM content <sup>a</sup>	GM variety <sup>b</sup>	GM %
Santa Maria	Taco shells	+	Bt176	<0.1
Santa Maria	Tortilla chips - chili - cheese Nachos chips	Tr	Category B	Tr
Snack-a-dip	Tortilla chips (+ salsa) - lightly salted - nacho cheese	+	Bt11	<0.1
St Michael	Authentic tortilla chips - ready salted - cheese flavour - chilli flavour	+	Category A	<0.1
Texicanos	Corn chips - cool ranch flavour - chilli flavour	+	Bt176	<0.1
Uncle Bens	Nacho chips Tortilla chips - plain	Tr	Category B	Tr
Wanted	Tortilla chips - salt - chili - cheese	+	NI	0.4

**Notes:**

- **No health or food safety concerns arise from these results; no companies were in breach of the GM labelling legislation<sup>1</sup>**
- a GM content:
  - ‘+’ GM material detected;
  - ‘-’ no GM material detected;
  - ‘Tr’ trace amounts of GM material detected
- b Where possible, GM varieties and % GM have been determined. However, in some cases, it was not possible to identify the actual GM variety, only that it is one of a group of varieties:
  - Category A = Bt176\*, Bt11\*, MON801, MON802, MON809\* and MON810\*
  - Category B = MON801, MON802 and MON810\*
  - \* Varieties approved for food use in the EU
- NI – Not Identified
- NA – Not Applicable

## 6. INTERPRETATION AND CONCLUSIONS

There are no known health implications arising from the presence of the GM ingredients identified in these food products. Only licensed GM ingredients that have already been subjected to a rigorous food safety assessment and approved for food use within the EU were positively identified in these foods.

GM maize ingredients were detected in 19 out of the 26 samples tested. Of those 19 positive samples, 8 contained only trace levels of GM DNA that was too low to be quantified. The remainder gave stronger signals, one contained 0.5% GM maize, another contained 0.4%, while all other samples contained <0.1%. The specific GM maize variety was identified in 5 samples, while a further 8 were established as containing maize from 1 of 2 categories of GM maize. Though 19 GM positive samples were detected, no unlicensed GM maize was positively identified in this survey.

The GM food labelling regulations require that foods containing GM ingredients at or above the 1% threshold must carry labelling to indicate that it contains GM ingredients. As all positive samples analysed in this survey contained levels of GM maize DNA below 1%, GM labelling was not required and therefore all the brands tested were compliant with the relevant legislation. However, the regulations stipulate that some form of identity preservation (or similar) system is in place to demonstrate that the presence of such GM ingredients are the result of adventitious contamination. Any Irish manufacturers of foods tested in this survey have satisfied the FSAI that there was no breach of this regulation.

At present, there is no specific European legislation relating to the use of labelling claims such as 'GM-free' and therefore, such labelling falls under the general requirements of the Food Labelling Regulations<sup>6, 7</sup>. Under these Regulations claims that are false or misleading to the consumer are prohibited. Therefore, a food containing GM ingredients, but labelled to indicate or suggest that it is GM-free may be in breach of this legislation. Although none of the foods sampled in this survey were specifically labelled as 'GM-free', one product bore the statement 'No Genetically Modified Corn Used' on the packaging. Analysis showed that the product in question contained <0.1% of a particular GM maize variety and thus, for the reasons outlined above, the labelling could be considered to be misleading.

The FSAI has contacted the retailers, suppliers and manufacturers whose products were included in the survey to inform them of the test results. Companies have also been notified of additional labelling issues and requirements, such as QUID with which they need to comply.

The FSAI will carry out further studies of selected foods containing soya or maize and is in the process of obtaining samples for analysis. This survey, and others planned for the future, constitute part of the FSAI's duty to ensure that only EU licensed GM foods are available on the Irish market and that such foods display the appropriate labelling information.

## APPENDIX A: FSAI SURVEY OF THE GM STATUS OF IRISH FOODS (1999/2000)

EU legislation governing the labelling of food products containing GM soya and GM maize was introduced in 1998.<sup>8</sup> To provide a broad overview of the Irish market with respect to the GM content of foods, the FSAI surveyed a range of food products to determine their GM status. The survey was not comprehensive and thus the results were not published but used as an indicator. The results of that survey are presented below, but due to the nature of the survey food types are listed instead of brand names.

Food Type	Relevant Ingredient(s)	GM present - Yes/No
Biscuits / cookies	- Soya bran - Bran, lecithin - Soya lecithin - Lecithin	No No No No
Bread	- Soya flour - Soya flour - Soya flour - Soya flour - Soya flour - Soya flour - Soya lecithin	No No No No No No No
Breadsticks	- Wheat flour	No
Burger buns	- Soya flour	No
Burgers	- Breadcrumbs - None apparent - None apparent	Yes* No No
Cereals / bars	- Corn starch - Maize flakes - Maize - Whole grain wheat - Whole wheat - Cornflour	No No No No No No
Chicken - southern fried - Kiev - breast fillets - nuggets	- Breadcrumbs - Breadcrumbs, wheatflour - Breadcrumbs - Starch, breadcrumbs, rusks, soya protein	No Yes Yes Yes
Chicken dishes - black bean sauce - Chinese	- Soy sauce, starch - Soy sauce, maize starch	No No
Cooked meats	- None apparent - None apparent	No No
Cooking sauces	- Not recorded - Tomatoes	No No
Crackers	- Wheat flour - Flour - Wheat flour	No No No
Cream cheese	- Soybean oil, tofu, soy protein, corn	Yes
Crisps	- None apparent - None apparent - None apparent - Corn flour	No No No No
Desserts (dairy)	- Cornflour	No
Dried milk	- Lecithin - Starch, lecithin	No No

<b>Food Type</b>	<b>Relevant Ingredient(s)</b>	<b>GM present - Yes/No</b>
Fruit cake	- Soya	No
Hot cross buns	- Lecithins	No
Infant milk/formulae	- Soy lecithin - Vegetable oils - Soy lecithin - Lecithin	No No No No
Lasagne	- Cornflour - Wheat protein, wheat flour, starch	No No
Lecithin granules	- Soya lecithin	No
Pasta bolognese	- Durum wheat semolina, starch	No
Pizza	- Maize starch, soya lecithin - Wheatflour, starch - Soya flour, starch - Wheatflour, starch - Wheatflour, starch	No No No No No
Popcorn	- Maize	No
Quiche	- Maize starch - Maize flour, maize starch	No No
Rusks	- Soya oil, lecithin	No
Sausage rolls	- Wheat flour, soya protein, starch, hydrolysed vegetable protein - Wheat flour, rusks, hydrolysed vegetable protein	Yes No
Sausages	- Soya protein, rusks - Rusk - Rusk - Rusk	Yes No No No
Savoury pies	- Hydrolysed vegetable protein (soya) - Wheat flour, starch, hydrolysed vegetable protein - Wheat flour, starch, hydrolysed vegetable protein - Wheatflour, maize starch, lecithin - Wheat flour, maize starch	No No No No No
Semolina	- Semolina	No
Soup	- Corn starch - Wheat, corn flour - Corn starch - Cornflour	No No No No
Soy sauce	- Soya flake - Soy beans	No No
Soya bran	- Soya bran	Yes
Soya cream/milk/yoghurt	- Hulled soya beans, lecithin - Soya protein - Soya beans - Soya beans and lecithin - Soya protein and lecithin - Soya beans	No Yes No No No No
Soya flour	- Soya flour	No
Spaghetti	- Durum wheat - Durum wheat - Durum wheat semolina	No No No
Spring rolls	- Cornflour, soy sauce, maize starch - Wheatflour, soy sauce, maize starch	Yes* Yes

Food Type	Relevant Ingredient(s)	GM present - Yes/No
Stuffing (fresh)	- Not recorded	No
Sweetcorn	- Sweet corn - Corn - Sweet corn	No No No
Taco shells	- Corn flour	No
Tortilla chips	- Maize - Corn - Maize flour	Yes Yes No
Vegetable and chicken dishes	- Cornflour - Soya flour	No No

**Notes:**

- \* denotes unauthorised GM ingredient detected
- It was not possible to identify the variety or quantify the amount of GM soya or maize present in any of these samples; the test was limited to distinguishing between approved and unauthorised GM ingredients.

The results of this survey show that 13 out of the 103 samples tested contained GM ingredients. Of this 13, none were labelled as containing GM ingredients and two were found to contain GM ingredients not approved for sale in the EU. One sample was labelled as containing vegetable protein produced from GM soya, but this was not detected in the sample, possibly due to the limit of detection of the analytical method available at that time.

These results revealed a general non-compliance with labelling regulations<sup>§</sup> which, at that time, required all foods containing detectable amounts of GM DNA or protein to be labelled as such. In addition, ingredients that were not approved for use in the EU were present in some foods sold in Ireland.

Companies whose products were identified as containing unlabelled GM ingredients were notified, and the two companies whose products contained unapproved GM ingredients were contacted with instructions to remove the products from sale.

**Note:** § – These regulations<sup>8</sup> have since been amended by the introduction of the 1% threshold for the presence of GM material in non-GM food ingredients, set by Regulation 49/2000/EC.<sup>1</sup>

## APPENDIX B: DESCRIPTION OF METHODS

The principal testing methodology employed is based on the extraction of DNA from food products (raw ingredients or processed food), followed by Polymerase Chain Reaction (PCR) analysis. This is designed to detect target sequences of DNA specific to Genetically Modified Organisms (GMOs).

**Sample Preparation:** All samples are homogenised to ensure that sub-samples taken for analysis are representative of the original sample.

**DNA isolation:** DNA is isolated from samples using a modified version of the CTAB method (*Article 35 of the Germany Federal Food Act, Method 24.01.1*). Some product categories are subjected to additional treatments to release the DNA from the sample matrix.

**Controls:** All samples analysed are extracted in duplicate, and each PCR analysed in duplicate to provide a confirmation of results. For each extraction, both positive and negative controls are used. The PCR analysis also has associated positive and negative controls. Positive controls are used to check the efficiency of DNA extraction, or the PCR analysis; negative controls, to ensure the absence of contamination and to avoid false positive results. The positive control is a certified GMO standard supplied by '*The Institute of Reference Materials*'.

As a means of excluding false negatives due to PCR inhibitors, samples in which no DNA is detectable are spiked with DNA, re-extracted and re-analysed. This ensures that the methodology employed is capable of detecting GM DNA if present in the product at the specified sensitivity.

**PCR Analysis:** Samples are initially screened for species DNA, e.g. soya (lectin gene) corn (*invertase* gene), oilseed rape (PEP Gene), etc., to determine if DNA can be detected in the sample. If DNA is detectable, samples are further screened for sequences which are characteristic of GM crops e.g., S-35, T-Nos, nptII, Bla, to look for GM-DNA - the exact choice of screening system depending on the species DNA found. Positive results from this initial screening are further confirmed using tests which screen for the specific genes used in GM crops – again the exact test used depends on the sample in question and the test requested, e.g. Cry1Ab, Cry1Ac, EPSPS, BAR, CaMV PolyA, Gox, Gus, or 'event specific' sequences.

Each sample is screened with at least 3 PCR tests, and all results have to confirm each other before a final result is reported. If results are inconsistent due to poor template quality, the sample is re-extracted and the whole screening process repeated.

**Detection:** PCR products are separated by conventional gel electrophoresis and visualised using Ethidium Bromide.

**Sensitivity:** The testing methodology can detect 10 copies or more of the target DNA sequences present in a sample. This is typically well below 0.01% GM DNA in a sample (as determined by international reference materials). The exact sensitivity of the detection system can vary from sample to sample.

**Validation:** The **qualitative GMO tests for processed food** have been validated by the Joint Research Centre of the EU.<sup>4</sup>

**Quantification:** An estimate of the amount of GM material is provided through using a Real-Time PCR analysis (Roche Lightcycler) and the Scorpion™ detection system.<sup>5</sup> Results are expressed as the percentage of GM DNA (as determined by S35\*) to Non GM DNA (as determined by species specific DNA, lectin, invertase, PEP...) in the sample. Both results are determined in a single analysis step (multiplex), overcoming errors due to each test being conducted in separate reaction mixtures.

*\* Use of the S35 system can over estimate the amount of GM DNA detected in a sample due to multiple copies of the S35 sequence found in particular GM 'events'. However, this more conservative approach is preferable to 'event' specific detection systems which underestimate the amount of GMOs in samples with multiple 'events'.*

## REFERENCES

1. Commission Regulation (EC) 49/2000, amending Council Regulation (EC) No 1139/98, concerning the compulsory indication on the labelling of certain foodstuffs produced from genetically modified organisms of particulars other than those provided for in Directive 79/112/EEC (Official Journal of the European Communities, L6 of 11.1.2000, p.13).  
**[[http://europa.eu.int/eur-lex/en/lif/dat/2000/en\\_300R0049.html](http://europa.eu.int/eur-lex/en/lif/dat/2000/en_300R0049.html)]**
2. Commission Regulation (EC) No 50/2000, on the labelling of foodstuffs and food ingredients containing additives and flavourings that have been genetically modified or have been produced from genetically modified organisms (Official Journal of the European Communities, L6 of 11.1.2000, p.15).  
**[[http://europa.eu.int/eur-lex/en/lif/dat/2000/en\\_300R0050.html](http://europa.eu.int/eur-lex/en/lif/dat/2000/en_300R0050.html)]**
3. 'General guidelines for implementing the principle of Quantitative Ingredients Declaration (QUID) - Article 7 of Directive 79/112/EEC as amended by Directive 97/4/EC', III/5260-rev5/98 - EN, 21 December 1998.  
**[[http://europa.eu.int/comm/food/fs/fl/fl02\\_en.pdf](http://europa.eu.int/comm/food/fs/fl/fl02_en.pdf)]**
4. EU examines accuracy of tests for genetically modified foods, British Medical Journal 2000, Vol. 320 pp 468 (Doug Payne, News Extra).  
**[<http://www.bmj.com/cgi/content/full/320/7233/468/d>]**
5. Detection of PCR products using self-probing amplicons and fluorescence, Whitcombe *et al*, Nature Biotechnology 1999, Vol. 17 pp 804-807.
6. Directive 2000/13/EC of the European Parliament and of the Council of 20 March 2000 on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs.  
**[[http://europa.eu.int/eur-lex/en/lif/dat/2000/en\\_300L0013.html](http://europa.eu.int/eur-lex/en/lif/dat/2000/en_300L0013.html)]**
7. European Communities (Labelling, Presentation and Advertising of Foodstuffs) Regulations, 2000, Statutory Instrument No. 92 of 2000 (Ireland).
8. Council Regulation (EC) No 1139/98 of 26 May 1998 concerning the compulsory indication of the labelling of certain foodstuffs produced from genetically modified organisms of particulars other than those provided for in Directive 79/112/EEC.  
**[[http://europa.eu.int/eur-lex/en/lif/dat/1998/en\\_398R1139.html](http://europa.eu.int/eur-lex/en/lif/dat/1998/en_398R1139.html)]**



## **FURTHER INFORMATION**

Further information on this survey can be obtained from:

Food Safety Information Centre  
Food Safety Authority of Ireland  
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