

June '04.

Report on F.L.E.P. Project – Official Control of ‘Gluten-free’ (G.F.) Foods.

Summary: 28 F.L.E.P. member states were forwarded an agreed questionnaire on Official Control of ‘Gluten-free’ (GF) Foods, and 12 useable replies were received. The relatively low response rate may somewhat limit this report’s use; however, the 12 respondents’ data are still of value, and are summarised below. Some variance is seen in the limits applied for gluten in ‘gluten-free’ (GF) foods, although generally 200ppm is the applied limit, and also, in some countries, 20ppm (for naturally GF foods) is used. Methods used in the Official Testing of foods are reasonably consistent, i.e. commercial ELISA, but there is a dearth of Proficiency Testing Schemes (PTS) and Certified Reference Materials (CRMs) for laboratories. There appears to be a large variance in both knowledge of gluten issues, and enforcement (surveillance and inspection) levels between responding member states. Nevertheless, most responding Food Control Authorities had some annual or occasional (‘special surveys’) monitoring/control mechanisms in place. Sampling of G.F. foods appears to be largely at retail level, although some countries emphasise production and other stages. Responses received on Official surveillance results for 2001 were good, with 9 countries able to produce “sample numbers tested” and “results ranges”. Considerable variance in Official testing levels exists. Overall, the rate of nominally G.F. foods exceeding 200ppm was 2.3% (ie 47 out of 2054). This rate appears high, in particular when one considers that the current Proposed Draft Codex upper limit of 200ppm is considered “questionable”^{1,2}. The results submitted also indicate that the majority of G.F. samples contain less than 50ppm gluten. The data indicate that manufacturers generally achieve levels of less than 50ppm gluten for both naturally G.F. and (wheat-based) foods rendered ‘gluten-free’. A number of recommendations are made, with a view to assist in improving Official Control of GF foods on the market.

1: Introduction.

Coeliac disease³ is an autoimmune disorder caused by an intolerance to dietary gluten in genetically predisposed persons. The coeliac condition has been described as being particularly prevalent in Scandinavia, the Celtic countries, and also in Italy; some recently quoted prevalence rates are close to 1 in 100 of the population. Medical experts describe the “coeliac iceberg”, where undiagnosed cases greatly outnumber diagnosed ones.

Gluten may be described as the proteinaceous mass remaining after washing wheat dough with water to remove starch. Chemically, gluten is composed largely of a complex, water-insoluble mixture of cereal storage proteins of varying molecular mass.⁴ The storage proteins of cereals include prolamins (‘ethanol-soluble’) and glutenins (‘ethanol-insoluble’). **Prolamins** are particularly high in the amino acids **proline** and **glutamine**. The prolamins of wheat, rye, barley and oats*

are gliadins, secalins, hordeins and avenins respectively. Other *Triticum* species of cereal, including spelt and kamut, also contain gluten, as do their crossbred varieties. The above cereals, except for oats*, are toxic, to varying degrees, to coeliacs.

**Some recent short-to-mid-term studies have shown that (pure) oats are not generally coeliac toxic, and Finland has recently⁵ requested the Codex Committee to consider deleting oats as a cereal not suitable for coeliac patients.*

The two main food groups considered safe for consumption by coeliacs are:

- (1): Naturally gluten-free (GF) foods
- and
- (2): Foods rendered 'gluten-free' (usually wheat-based).

The presence of gluten in foods nominally gluten-free may be regarded as a contaminant (for naturally 'gluten-free' foods) or as a residue (in foods rendered 'gluten-free').

Thus the issue of gluten in supposedly gluten-free foods, being one of food safety, labelling and trade (single EU market), may be dealt with as an Official Control of Foodstuffs matter. Because of the above, the subject was developed into a F.L.E.P. (Food Law Enforcement Practitioners)⁶ project, in particular as there is no specific EU legislation on limits for gluten in 'gluten-free' foods.⁷ A Proposed Draft Codex Standard (step 7) for gluten in 'gluten-free' foods is under discussion by a Codex Committee on Nutrition and Foods for Special Dietary Uses (CCNFSDU). Limits of 20ppm (naturally G.F. foods) and 200ppm (foods rendered 'gluten-free') are proposed, but these are not likely to be adopted before tolerable intake studies and food analytical method approval (by Codex CCMAS) are completed (possibly by end of 2004 or 2005?).

Food Control Authorities participating in these Codex deliberations on standards for gluten-free foods require a knowledge of gluten toxicology/clinical sensitivity issues, and also they must also address the critical issue of whether the limit(s) are to be set in conjunction with a specified test method(s), or whether the method is to be discretionary.

Much of the work on establishing reliable methods of analysis and tolerable intakes has been driven by the Working Group on Prolamin Analysis and Toxicity (P.W.G.)^{7A}. Other valuable research in these areas continues elsewhere, generally in hospital and university centres.

2: F.L.E.P. Project – Official Control of 'Gluten-free' Foods.

At the FLEP forum in Cork in 2002, a proposal by Ireland to gather basic comparative information on how member states monitor/control gluten in 'gluten-free' foods was accepted. A number of countries agreed to participate in a working group⁸ set-up to report on collated information gathered from member states (via FLEP contact points). The meeting agreed to conduct the project by E-mail questionnaire with Ireland as principal coordinator. A draft questionnaire was submitted to 10 working group countries via F.L.E.P. contact points, including EU member states and other FLEP participants.⁶ An agreed questionnaire (see Appendix A) was forwarded in May 2002 to 17 FLEP contact points^{8a} or to their nominated national expert. In June 2002 the agreed questionnaire was e-mailed to 3

other Contact Points⁹ and posted to 8 others¹⁰ (and re-posted to same 8 in July 03). 7 Countries¹¹ submitted prompt replies to the Questionnaire by Sept. '02. A further 6¹¹ replies were received up to Oct 2003, at which point it was decided to write-up a summary report of 12 usable responses,¹² despite the relatively low number of responded Questionnaires received (13 out of 28 countries requested).

3: Summary of Results.

Results received from 12 countries are collated for Questions 1-6 of the Questionnaire - see Appendices 1 to 5 respectively for tabulated summaries of national responses.

3.1: National Limits.

Q.1.1. “Does your member state (MS) prescribe national limits or guidelines for gluten in each/any of the following products: (if yes, please specify limit)?”

Responses: (see Appendix 1 for summary of results).

The majority of respondents (8 out of 12) stated that their country does not have prescribed limits. Four countries, Switzerland, Sweden, Czech Republic and the Netherlands, stated that they had national prescribed limits in force.

Of the four, only Sweden has a prescribed limit (a guideline) of 20ppm for naturally G.F. foods.

Q.1.2. “If answer(s) to 1.1 above is No, what limits/acceptance criteria are generally applied in your M.S. to foodstuffs tested for gluten?”

The responses to this question indicate that of the 8 member states without national prescribed limits (for gluten in ‘GF’ foods), 7 apply limits of 200ppm (or 20ppm and 200ppm) in practice. Therefore, overall, 11 of the 12 responding MSs appear to have useable acceptance criteria for gluten in GF foods; the above appears to be true both for naturally G.F. foods and for foods rendered ‘gluten-free’.

The reported variance in “National Limits” could potentially lead to cross-border food law enforcement problems, although to date few such problems appear to have arisen.

3.2. Methods of Analysis.

Q.2.1. “What methods of analysis are generally used in your MS for testing Gluten in Foods?”

Q.2.2. “Is an additional method used to confirm ‘high’ results... if so, give brief outline of method.”

Q.2.3. “Does the laboratory (a) generally use a CRM (or ‘Quasi’ CRM etc)?

or (b) participate in a Proficiency Test or Ring Test?”

Responses: (see Appendix 2 for summary)

Generally commercial ELISA kits are used routinely. Four countries use confirmatory methods (PCR, 'Other' ELISA kit, MS, etc) for 'high'-gluten samples. Respondents do not use matrix-matched CRMs, eg GF flours or starches, as none are available (Note: the new European gliadin standard^{12A}, IRMM-480, first produced by the Working Group on Prolamin Analysis and Toxicity (PWG), is used for calibration by some Member State labs; this standard is currently (May '04) being certified by IRMM (EU)).

Note: some ELISA kit manufacturers now base their method on calibration using the new European gliadin reference material^{12A} and antibodies/extraction procedure developed by Dr. Mendez of P.W.G.^{7A}

3.3 + 3.4. Official Foodstuffs Tested and Sampling Stage/Quantity.

Q.3.1. "Approximately how many official food-control samples from the following Categories of food are tested per annum in your member state?"

Q.4.1. "Please give brief details on sampling stages and sample quantities..."

Responses (see Appendix 3 for summary).

Two countries of the 12 responded that Official testing of G.F. foods is not carried out. Ten of the 12 generally perform either routine, programmed annual testing or perform occasional special surveys (or a mixture of both!). As might be expected, a substantial variation in the level of Official surveillance can be seen, with e.g. a relatively high number of samples being tested in Germany, - see also Section 3.6 below covering 2001 Surveillance Results.

Sampling stage is generally "largely retail", although some respondents indicate that manufacturing and other stages are also included. Respondents generally indicated that one commercial packet is usually taken for analysis.

3.5. Inspections, etc.

Q.5.1. Number of G.F. Manufacturers, etc. in your Member State? (& No. of Inspections per annum, per premises).

Q.5.2. Number of 'G.F.' Bakeries in your Member State?(& No. of Inspections per annum, per premises).

Q.5.3. In above Inspections (Q.5.2.) is 'Gluten' HACCP monitored?

Q.5.4. Are there National/Regional guidelines available for 'Gluten Control' in G.F. foods?

Responses (see Appendix 4 for summary).

Most MS were able to provide information on their number of G.F. Producers, although information on the number of G.F. bakeries was less readily available (6 out

of 12 countries provided the figures). Where available, the information provided indicates that each G.F. 'premises' is inspected generally between once and twice per annum. Only 4 of the 12 respondents stated that the HACCP control points related to gluten are monitored

Only one country's respondent (Sweden) indicated that national/regional guidelines were available in their MS for controlling gluten in G.F. foods. One country's (Ireland's) Food Safety Authority has recently prepared such Draft Guidelines.¹³

3.6: Results of Analysis and Food Complaints.

Q.6.1. Summary of National Test Results for Gluten in G.F. Foods for 2001.

Responses: (see Appendix 5 for summary).

Of the 12 responding MS, 9 provided data on total 'gluten-free' sample numbers tested (for gluten) in 2001. Of the 2054 Official samples reported as tested in 2001, 47 (ie 2.3%) exceeded 200ppm, the upper, proposed Codex limit.

It is important to interpret the tabulated data (see Appendix 5) with caution, in particular as they include both naturally G.F. and wheat-based foods rendered 'gluten-free'; also the 47 samples with gluten above 200ppm would be expected to include a proportion of targeted, 'follow-up' and 'food complaint' products. Another reason for caution is that the results of analysis are to quite an extent method dependent, due largely to the complexity of the parameter (gluten) being determined. Nonetheless, the results indicate a significant number of 'excessive gluten' samples, in particular when considered in the context of the Codex proposed upper limit of 200 ppm being "questionable"^{1,2}. Although not quantifiable from the data received, a significant number of the tested naturally G.F. foods can be expected to be in excess of 20ppm, the currently Proposed Draft Codex limit (see Appendix 6 for a breakdown of results from 1 laboratory).

On a positive note the results indicate that manufacturers generally achieve gluten levels of less than 50ppm for the majority of G.F. products, both naturally G.F. and rendered 'gluten-free'. Data from Ireland¹⁴ indicate that 86% of tested wheat-based foods rendered 'gluten-free' contain gluten levels less than 50ppm, indicating that manufacturers generally achieve levels well below the proposed, upper Codex limit of 200ppm! (see also Appendix 6).

Note: during the course of collecting the above results, the coordinator was provided with data on extensive testing of GF foods in Spain (in Dr Mendez's laboratory, Madrid). Although the data differ from those discussed above in that they are not the results of Official monitoring, they are worth mentioning here because of the large number of samples tested, and due to the fact that the results^{13a} were generated in the laboratory which developed the new ELISA R5 procedure. Of the 4454 samples tested, 347 (ie 7.8%) exceeded 200ppm.

Also, the Swedish participant reported that "...a private laboratory in Sweden...performs about 1000 analyses per year" (see Appendix 3, Note 1). Other respondents did not supply this additional information but it is likely that testing by private laboratories also takes place in other member states.

Food Complaints concerning G.F. Foods.

Q.6.2 “Provide general information of investigations of foodstuffs linked to reported illness in coeliacs...”

Responses: (see **Appendix 5 for summary**) On this difficult-to-answer subject, 6 member states provided information, with a comprehensive response from Sweden (see Appendix 5a). Overall the number of reported (to the FLEP Questionnaire) annual cases of acute adverse reactions is low. Of note are the Swedish data, where 4 of the 11 cases reported yielded gluten levels of less than or equal to 200ppm in the suspected offending food (see Appendix 5a).

4: Recommendations.

1: That Gluten be recognised by all EU Member States, including new entrants, as an important allergen, requiring a sustained, proportional food control mechanism applied by both Industry and National Official Food Control Agencies. The above mechanism should include, as appropriate, inspection/audit and surveillance, in particular to include Manufacturing (including Bakery), Retail and Import Stages.

2: That in addition to foods rendered ‘gluten-free’, infant foods and baby foods, and other naturally ‘gluten-free’ foods (including those listed in National Coeliac Societies’ Books of “permitted” foods) be subject to the control referred to in 1 above.

3: That the EU Commission facilitate 1: the IRMM (and other agencies where appropriate) to provide a broad range of suitable G.F. CRMs, eg GF starches, flours, for validation use by testing laboratories, and 2: the IRMM and other appropriate agencies, eg CSL(FAPAS) and other PTS scheme providers, to provide a suitable range of Proficiency Testing Schemes for laboratories.

4: That laboratories recognise the complexities of the test parameter “gluten” and the difficulties with testing heat-treated and (partially) hydrolysed samples. Labs should always ensure that they employ the most appropriate extraction and detection techniques for each GF food type tested. The new R5 Sandwich ELISA method¹⁵ using Mendez Cocktail Solution as extraction solvent (for heat-treated GF foods), and a new competitive R5 ELISA¹⁶ (for (partially) hydrolysed GF foods), represent important recent advances in solving some of the problems of gluten analysis in foods. Laboratories should keep abreast of relevant developments concerning international validation/approval of gluten test methods by Codex CCMAS, AOAC etc.

EU member state laboratories should use analytical techniques which generate demonstrably comparable quantitative results. See also Note in Appendix 6.

References etc.

1: Analysis and Clinical Effects of Gluten in Coeliac Disease, M. Stern *et al*, Eur. J. Gastroenterol & Hepatol 2001, 13, p. 741-747.

- 2: Report of the Working Group on Prolamin Analysis and Toxicity, Codex Committee on Nutrition and Foods for Special Dietary Uses – CX/NFSDU 03/4 – Oct. 2003.
- 3: See eg. Coeliac Disease(a clinical review), Feighery C, BMJ, 1999; 319:236-239; Ciclitara P, Gastroenterol 2001, 120, p. 1526-1540; also Ref. 1 above.
- 4: See eg. Weiser H, Cereal Protein Chemistry. In: Feighery C, O’Farrelly C(Editors), Celiac Symposium, Dublin 1992. Dublin: Oak Tree Press, 1994, p. 191-202.
- 5: Codex Alimentarius Commission (ALINORM 04/27/26 par. 31 and CRD 5 from the 25th session of CX/NFSDU) Codex Committee on Nutrition and Food for Special Dietary Uses; Proposed Draft Revised Standard for Gluten-free Foods at Step 7, - Germany, 3-7 Nov. 2003 (See comments from Finland).
- 6: See F.L.E.P. Constitution – F.L.E.P. Bulletin July 1998.
F.L.E.P. Website: <http://www.flep.org>
- 7: However, infant formulae should not contain any source of gluten (see Commission Directive 91/321/EEC and subsequent amendments). Also, processed cereal-based foods and babyfoods for infants and young children must declare the presence or absence of gluten on the label (see Commission Directive 96/5/EC and subsequent amendments).
- 7A: The Working Group on Prolamin Analysis and Toxicity (P.W.G.) is a multi-disciplinary, medical and scientific, group working largely on co-ordination of research into gluten analysis in food and on clinical evaluation of coeliacs’ sensitivity to gluten. P.W.G. has “observer” status at Codex meetings (on gluten limits, etc.). Bound Reports of PWG’s annual meetings are produced and these form an excellent source of information on gluten analysis and toxicity.
- 8: The FLEP Working Group volunteered members: Ireland, Netherlands, U.K., Germany, Denmark, Finland, Latvia, Czech Republic, Austria and France; Dr. Stroka (EU Reference Lab.) also expressed interest.
- 8a: See list of Contact Points(with E-mail addresses) - 2002 , FLEP Secretariat.
- 9: Lithuania, Greece and Bulgaria (e-mail addresses provided by FLEP secretariat).
- 10: Iceland, Italy, Luxembourg, Portugal, Spain, Poland, Romania and Slovak Republic (e-mail addresses not available).
- 11: The tabulated data in the Appendices are in order of receipt of responded Questionnaires from participants.
- 12: See Appendices 1 – 5 for responding countries.
- 12A: See reference 5, Appendix 2.

13: As part of the work of the Food Safety Authority of Ireland's (F.S.A.I.'s) Working Group on Gluten, Draft Guidelines on avoiding cross-contamination during preparation of Gluten-free foods have been prepared; these are aimed at both industry and at official control personnel.

13a: the test results provided were as follows: results range : <3ppm gluten:1604 samples; 3-20ppm: 1209 samples; 20-100ppm:1079 samples; 100-200ppm:215 samples; >200ppm: 347 samples. Results were obtained using the ELISA R5 method.

14: Of the 289 official samples of (wheat-based) foods rendered 'Gluten-free' tested in 2001 – 2003 inclusive, 249 (86%) contained less than or equal to 50 ppm gluten. These results were generated in the Public Analyst's Laboratory, Western Health Board, Galway, Ireland, using ELISA methods (r-Biopharm Ridascreen Gluten and R5 Ridascreen Gliadin).

15: See Reference 6, Appendix 2.

16: Ferre S et al. (2003), Measurement of hydrolysed gliadins by a competitive ELISA based on monoclonal antibody R5: analysis of syrups and beers, Stern M (Ed.), Proceedings of the 18th Meeting of Working Group on Prolamin Analysis and Toxicity, Oct. 2003 Stockholm, Sweden; Verlag Wissenschaftliche Scripten, Zwickau, 2004, Germany, p. 65-70.

Appendix A

FOLLOWING THE FLEP MEETING – CORK, IRELAND 4th & 5TH FEBRUARY 2002

AGREED QUESTIONNAIRE ON OFFICIAL CONTROL OF 'GLUTEN-FREE' MARKET FOODSTUFFS

LIMITS FOR 'GLUTEN-FREE' FOODS:

1.1: Does your member state (MS) prescribe national legislative limits or guidelines for gluten in each/any of the following products: (If yes, please specify limit):

(a): naturally 'gluten-free' foods	-	<u>Yes/No :</u> _____	_____ mg/kg
(b): wheat-based, deglutenised foods	-	<u>Yes/No :</u> _____	_____ mg/kg
(c): infant foods	-	<u>Yes/No :</u> _____	_____ mg/kg
(d): other foodstuffs	-	<u>Yes/No :</u> _____	_____ mg/kg

Comments (if required):

1.2: If answer(s) to 1.1 above is No, what limits/acceptance criteria are generally applied in your MS to foodstuffs tested for gluten?

Answer

- (a): naturally 'gluten-free' foods: _____ mg/kg
(b): wheat-based, de-glutenised foods: _____ mg/kg
(c): infant foods: _____ mg/kg
(d): other foodstuffs: _____ mg/kg
Comments (if required):

METHOD(S) OF ANALYSIS:

2.1 What methods of analysis are generally used in your MS for testing gluten in foods?

Answer:

Note: if a commercial ELISA procedure is used, please give kit name and Manufacturer's name.

2.2 Is an additional method used to confirm 'high' results from first method? If so, please give brief outline of method.

Answer:

2.3 Does the testing laboratory (a) generally use a CRM (or 'Quasi' CRM etc)*?

Answer:

or (b) participate in a Proficiency Test or Ring-Test*?

Answer:

* If yes, please give name/details.

Comments (if required) :

FOODSTUFFS TESTED:

3.1 Approximately how many official food-control samples from the following categories of food are tested per annum in your MS?

- (a): naturally 'gluten-free' foods: _____ tested per annum
(b): wheat-based, de-glutenised foods: _____ tested per annum
(c): infant foods: _____ tested per annum
(d): other foodstuffs: _____ tested per annum

Comments (if required):

SAMPLING/ SOURCES:

4.1 Please give brief details of the stages of sampling of gluten-free products in your MS. - eg. use the scheme below to answer this question.

Answer:

- (a) Solely retail level
(b) Largely retail
(c) Largely manufacturing/processing
(d) All stages

4.2 Give brief details on sample quantities generally taken for gluten analysis.

Answer:

INSPECTIONS ETC:

5.1 How many manufacturers or processors (excluding small bakeries) produce gluten-free products in your MS.

Answer = []

- How many 'gluten control' inspections are carried-out per annum (per premises) on these 'producers? Answer = []
- 5.2 How many bakeries produce 'gluten-free' products in your MS?
Answer = []
How many 'gluten-control' inspections are carried-out per annum(per premises) on these Bakeries?
Answer:
- 5.3 During inspections of above premises are the gluten-related HACCP control-point(s) monitored?. Is the potential for cross-contamination monitored?
Answer:
- 5.4 Are there national/regional guidelines available in your MS for 'controlling' gluten in 'gluten-free' foods?
Answer:
- Comments (if required):

RESULTS OF ANALYSIS:

- 6.1 Can you provide a summary of your MS's results of analysis of gluten, in 'gluten-free' foods for 2001, for all 'gluten free' samples tested:

2001	No. of Samples	Results Range
		0-50mg/kg
		50-100mg/kg
		100-200mg/kg
		>200mg/kg

Comments (if required):

- 6.2 FOOD COMPLAINTS CONCERNING 'GLUTEN- FREE' FOODS:** can you provide general information of any investigations of foodstuffs linked to reported illness in coeliacs in your MS?

e.g. <u>Sample Type/Description</u>	<u>Number(s) affected</u>	<u>Results of Analysis</u>
Wheat-based, 'gluten-free' white flour.	1 x 15- year old boy	600 mg gluten/kg

Answer:

Comments (if required):

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Appendix 1.

Q.1: Limits for ‘Gluten-free’ (G.F.) Foods.

	Q.1.1. : Prescribed National Limits or Guidelines				Q.1.2: Other Limits/acceptance criteria applied...			
Country	Naturally G.F. Foods	Foods rendered ‘Gluten-free’	Infant Foods	Other Foods	Naturally G.F. Foods	Foods rendered ‘Gluten-free’	Infant Foods	Other Foods
Latvia	No	No	No	No	200ppm	200ppm	200ppm	200ppm
Switzerland	No	200ppm*	No	No	--	See left	--	--
Sweden	20ppm	200ppm	No	No	see left	See left	20 or 200ppm ¹	20 or 200ppm ¹
Germany	No	No	No	No	200ppm	200ppm	200ppm	200ppm
Norway	No	No	No	No	20ppm	200ppm	20 or 200ppm ¹	20 or 200ppm ¹
Czech Republic	200ppm	200ppm	200ppm	200ppm	See left	See left	See left	See left
Luxembourg	No	No	No	No	200ppm	200ppm	200ppm	200ppm
Denmark	No	No	No	No	--	--	²	--
Netherlands	No	200ppm	No	No	200ppm ³	See left	200ppm ³	200ppm ³
Malta	No	No	No	No	200ppm	200ppm	200ppm	200ppm
Spain	No	No	No	No	--	--	--	--
Ireland	No	No	No	No	20ppm	200ppm	20 or 200ppm ¹	--

* Specifically: 100mg prolamin(gliadin)/kg dry material.

¹20ppm if naturally GF, 200ppm otherwise.

²“Must not contain gluten....- infant formula”.

³“Only where product claims to be gluten-free”

Appendix 2: Q.2 Methods of Analysis.

Country	Q.2.1: Method	Q.2.2: Add'l Method for Confirmation	Q.2.3.a CRM use	Q.2.3.b Proficiency testing/ Ring Test.
Latvia	No testing	Not applicable	Not applicable	Not applicable
Switzerland	ELISA (Transia Plate Gluten)	No	No	No
Sweden	ELISA (Transia Plate Gluten)	PCR	PWG Std. ⁵	Yes-FAPAS and PWG ⁶
Germany	ELISA (r-Biopharm.. ¹) etc.	In 2 labs. ²	In 1 lab. ³	Yes (LVU ⁴ ...)
Norway	ELISA (Tepnel Biosystems..)	No	---	Yes(“Ring Test”)
Czech Republic	ELISA (Ridascreen-r Biopharm)	Yes (ELISA Riedel de Haen <u>or</u> ELISA TEC)	Kit Starches only	No
Luxembourg	ELISA (Ridascreen..R-Biopharm)	No	Kit Standard Only	“Not yet”
Denmark	No testing	Not Applicable	Not Applicable	Not Applicable
Netherlands	SDS-Electrophoresis + Immunological Method.	No	No. (In-house Standard Control)	Yes (FAPAS-UK)
Malta	ELISA (Ridascreen Gluten Kit R6101)	No	No	--
Spain	ELISA R5(Inginasa and r-Biopharm) and Western Blot	PCR	IRMM-480 ⁵ And Ref Material 8418 NIST,US ^{5A}	No
Ireland	ELISA (R-5, r-Biopharm or Tepnel Biosystems)	Ocassionally only. (ELISA in External Labs.)	PWG Std ⁵ .	FAPAS (UK).

NB SINCE RECEIVING SOME OF THE ABOVE RESPONSES, SOME ELISA KIT MANUFACTURERS HAVE BROUGHT OUT NEW KITS; THEREFORE SOME OF THE ABOVE REPORTED METHODS MAY NOT BE FULLY UP TO DATE NOW(MAY '04).

¹ Also ELISA-Immunolab GMBH; Diffchamp/Transia; ELISA Corimg and immunodiffusion by Ochterlony (in Berlin Lab.)

² PCR Confirmation in 2 labs. (generally not confirmed).

³ IRM (generally no participation).

⁴ “Mostly Yes: Proficiency Test LVU Lippold, D- 79336 Herbolzheim.”

⁵ PWG Std = European Gliadin Reference Material, first produced by Prolamin Working Group..., now available as IRMM-480 from IRMM (EC Institute for Reference Materials and Measurements), Directorate-General Joint Research Centre (JRC), Reference Materials Unit, Retieseweg, 2440 Geel, Belgium.

- see Van Eckert R (2002): The PWG gliadin, a new reference material, in Stern M (Ed.): Proceedings of the 16th Meeting, Working Group on Prolamin Toxicity and Analysis, Nov. 8 – 11, 2001, Sitges, Spain. Verlag Wissenschaftliche Scripten, Zwickau, Germany, p. 25 – 27.

^{5A} Reference Material No. 8418(Wheat Gluten)- US Dept. of Commerce, National Institute of Standards and Technology, Gaithersburg MD 20899, USA.

⁶ Immer U et al (2003), PWG collaborative trial of gluten-free food through “Cocktail ELISA”, Stern M (ed.), Proceedings of 17th Meeting of Working Group on Prolamin Analysis and Toxicity, Oct. 2002, London, UK; Verlag Wissenschaftliche Scripten, Zwickau, Germany, p. 23 – 33.

Appendix 3: Qs. 3 & 4 Foodstuffs tested and Sample Sources.

Q.3.1 Foodstuffs Tested Annually					Q.4 Sampling Sources, etc.	
Country	‘Naturally’ G.F. Foods	Foods rendered GF	Infant Foods	Others	Q.4.1 Stage	Q.4.2 Quantity
Latvia	No annual testing generally				Not Applicable	Not Applicable
Switzerland	--	40	--	10	Solely retail level	1 commercial packet
Sweden	(Note 1)	(Note 1)	(Note 1)	(Note 1)	Largely retail	1 packet
Germany	200	50	450	750	All stages	Normally 1-3 packets..
Norway	156 (combined)		0	79 (Note 2)	Largely retail	Generally one sample.
Czech Republic	25	15	5	20	All Stages (Note 3)	--
Luxembourg	10	4	2	4	Largely retail	Normally >100g.
Denmark	No annual testing generally				Not applicable	Not applicable
Netherlands	--	50 (Note 4)	--	--	Largely retail	One unit
Malta	0	7	6	1	Import/Retail	Unit Packet
Spain	95	20	41	45	Solely Retail	--
Ireland	ca. 300	ca. 150	ca. 40	0	Largely retail	One packet

Note 1: Routine programmed testing is not usually carried out. However some projects are carried out, eg. 1997/8: 31 samples tested (EU Co-ordinated Programme). In 2002 gluten-free foods were tested as part of a “special medical purposes” project. “In Sweden a great number of analyses have been performed by the authority as well as by private laboratories on contract as a customer service but not as official control”. “...a private laboratory in Sweden...performs about 1000 analyses per year”.

Note 2: These figures refer to 2000/2001. Regular annual monitoring is not performed.

Note 3: “The rate is: Manufacturers: Retail = 2:1”.

Note 4: In 2001 more extensive testing was performed.

Appendix 4

Q. 5 Inspections, etc.

Country	Q.5.1 No. of G.F. 'Producers'	Q.5.1 No. of Inspections per Producer	Q.5.2 No. of G.F. Bakeries	Q.5.3 No. of Inspections Per Bakery	Q.5.3 Are G.F. HACCP Pts 'Inspected' ?	Q.5.4 National/ Regional Control Guidelines
Latvia	1 (Note 1)	--	--	--	--	--
Switzerland	"0"	Not applicable	4	1	No	No
Sweden	ca. 44	Note 2	Ca. 27	(Note 2)	Yes	Yes
Germany	ca. 19	2	10	1.8	Yes	No
Norway	Not available	Not available	Not available	Not available	Not available	No
Czech Rep.	ca. 25	ca 1.24	Ca. 10	ca. 1.5	Yes	No
Luxembourg	None	Not Applicable	6	1-2 (Note 3)	No	No
Denmark	Not available	Not available	Not available	Not available	Not available	Not available
Netherlands	ca. 10	1 minimum	Not available	Not available	Yes	(Note 4)
Malta	0	Not Applicable	0	Not Applicable	Not Applicable	No
Spain	--	--	--	--	--	--
Ireland	6	ca. 2				Yes (Note 5)

Note 1: One G.F. bread producer in Latvia (separate production line used). Producer collaborates with Baltic Celiac Association.

Note 2: No routine number; -see Note 1, Appendix 3.

Note 3: 1 – 2 from 2002 (in 2001 : 8).

Note 4: "Guidelines of the Codex Alimentarius".

Note 5: "Such guidelines are in Draft Form and have been prepared (Mar '04) by the Food Safety Authority of Ireland's (FSAI's) working group on gluten.

Appendix 5.

Q.6: Results of Official Analysis.

Q.6.1 2001 Official Results (ppm Gluten).					Q.6.2 Complaints re G.F. Foods
Country	0-50	50-100	100-200	>200	
Latvia	No testing	See left	See left	See left	None received (by State Food/ Veterinary Service).
Switzerland	35	3	2	9	Not available
Sweden	See Note 1				Detailed data (see Note 3 below)
Germany	1104	18	156	12 Note 1A	“Not available”
Norway	30 (41)	3 (4)	3 (1)	0 (6) (Note 2)	?
Czech Rep.	37	12	10	6	7 in 2001 (legislative limits exceed in 2 of these cases!)
Luxembourg	30	0	0	1	“None known of”
Denmark	No testing	See left	See left	See left	Not available.
Netherlands	90			7	“About 5 complaints per annum”
Malta	-(Note 4)	--	--	--	No complaints received
Spain	167	19	6	9	--
Ireland	263	11	8	3	Estimated 2-5 per annum

Note 1: Gluten Projects carried out in 1997/98 and in 2002 (not in 2001).

Note 1A: “6 samples (of the 12) from same manufacturer.”

Note 2: “Numbers in brackets for products where flour is not in ingredients list.”

Note 3: 12 cases seen since 1992 (see Appendix 5A for breakdown).

Note 4: “All results <0.02% gluten” gluten (no breakdown given).

Appendix 5A.

Swedish National Food Administration.
Dr. Ingrid Malmheden Yman

Adverse reactions to gluten

Consumers' complaints over adverse reactions have led to analysis of the suspected offending food and documentation of these cases. In total we have since 1992 seen 12 cases, where consumers have reacted.

Food (Month/year)	Reaction	Content Gluten (ppm)*	Cause
Buckwheat flour (05/92)	Stomach pain, diarrhoea <i>Adult with coeliac Disease</i>	13000	15-25% wheat flour identified, may emanate from the mill
Corn Pasta (02/92)	Allergic reaction <i>Child with allergy Against wheat, rye, egg</i>	3000	Contamination
“Gluten free” pasta (02/96) (Case 1)	<i>A boy, 9 years old, with Coeliac disease, is vomiting At three occasions after having eaten the pasta</i>	10000	Contamination of the raw material or during production
(Case 2)	<i>A girl, 8 years old, with an allergy to cereals suffers at three occasions from breathing difficulties and asthma after having eaten the pasta at school lunch</i>		
“Gluten free” pasta (spirals) (11/96)	Stomach pain, vomiting <i>Child with coeliac disease</i>	200	Contamination during production Gluten content was 5% in one piece of “band” pasta found in the package
Bisquits, glutenfree (03/96) (Case 3)	Stomach pain, skin symptoms <i>Adult with coeliac disease</i>	140	Contamination during production
Potato croquettes (07/93)	Stomach pain <i>Adult with coeliac disease</i>	900	The manufacturer had “forgotten” to declare the ingredient wheat flour
Snacks (03/97) (Case 4)	Vomiting <i>Boy, 6 years old with coeliac disease</i>	10000	The ingredient (gluten or wheat flour) was not declared.
Chocolate (12/95)	Stomach Pain <i>Child with coeliac disease</i>	4200	Misleading declaration. The ingredient label says rice crisps, the picture shows wheat crisps
Chocolate bar with rice crisps (11/94) (Case 5)	Stomach pain, diarrhoea <i>Adult with coeliac disease</i>	70	The rice had been treated with gluten, this was not declared
Wafer (01/97) (Case 6)	Stomach pain <i>Child, with coeliac disease</i>	70	Naturally gluten free product, Contaminated with wheat gluten
Ice cream cone	Vomiting	860	Falsely labelled as gluten free

* The results were obtained with the Transia gluten assay, based on monoclonal antibody to omega gliadin.

Appendix 6

Gluten testing in Public Analyst's Laboratory, Galway – Results for 2003

Food category	Limit Applied (ppm)	No. of Samples Tested	Results Range (ppm)				
			<20	21 – 50	51 - 100	101 – 200	>200
Foods rendered 'Gluten-free'	200	108	64	27	8	1	8
Naturally gluten-free products	20	261	202	16	21	14	8
TOTALS		369	266	43	29	15	16

All the 2003 samples above were tested using the R-Biopharm R5 ELISA,- Ridascreen Gliadin. This method was further improved during the course of 2003, with the introduction of the Mendez cocktail solution which is now used for all samples. This improves the detection of the prolamin fractions in heat-treated products. Samples were Official ones submitted largely by Environmental Health Officers.

Note: the following comment was provided by the Swedish participant:
“According to the provider R- Biopharm, the cocktail extraction gives false positive results in certain food matrixes like those containing milk. Ethanol should be used unless heat-processed samples are being analysed”.