Food Allergens: Challenges and developments
Michael Walker EHAI/CIEH Conference 21 May 2015

Science
for a safer world
## LGC – a global company

<table>
<thead>
<tr>
<th>Year</th>
<th>Event</th>
<th>Employees</th>
<th>Turnover</th>
</tr>
</thead>
<tbody>
<tr>
<td>1842</td>
<td>Laboratory of the Board of Excise founded to protect excise duty payable on tobacco importation into the UK, became Laboratory of the Government Chemist with technical appeal functions</td>
<td>270</td>
<td>£15m</td>
</tr>
<tr>
<td>1996</td>
<td>Laboratory of the Government Chemist privatised</td>
<td>678</td>
<td>£56m</td>
</tr>
<tr>
<td>2003/04</td>
<td>Focus on science-dependent activities</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2009/10</td>
<td>Acquisitions focused on services (Agowa, Forensic Alliance)</td>
<td>1,380</td>
<td>£130m</td>
</tr>
<tr>
<td></td>
<td>Over 35% of revenue within Standards, the only product business</td>
<td></td>
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<td></td>
<td>Footprint across Europe (focused on UK and Germany)</td>
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<td></td>
<td>50% revenue on products, with KBio acquisition and Standards growth</td>
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<tr>
<td></td>
<td>Increasing footprint in US and RoW</td>
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<tr>
<td></td>
<td>2,000 employees</td>
<td></td>
<td>£200m</td>
</tr>
<tr>
<td></td>
<td>£200m turnover</td>
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</tbody>
</table>

**Today**

Focus of activities is on intellectual property and new product development

- Acquisitions focused on pharma and agbio (KBio in Genomics, QBAS in Health Sciences)
- Over 35% of revenue within Standards, the only product business
- Footprint across Europe (focused on UK and Germany)
- 50% revenue on products, with KBio acquisition and Standards growth
- Increasing footprint in US and RoW
- 2,000 employees
- £200m turnover
LGC
Laboratory of the Government Chemist

Standards
Genomics
Laboratory and Managed Services

Science & Innovation
Group Functions
LGC’s UK national roles

National Measurement Institute (NMI)

- The UK’s designated NMI for chemical and bioanalytical measurement
- Provides traceable and accurate standards of measurement for use in industry, academic and government

Government Chemist

- Referee Analyst
- Adviser to government and industry on regulations & scientific input
- Allergen measurement research

“Using sound analytical science in support of policy and regulation”
Aims

✓ Improve quality of life
✓ Make it easier for food companies to supply safe food
✓ Promote exchange of knowledge about all aspects
✓ Over 650 members

http://safefoodallergy.ning.com/

3 videos showing allergen management
In a busy hotel kitchen –on YouTube
http://www.youtube.com/watch?v=PTBI3hCPT94&list=PLh7qvGGhPbb0T0fpY8KVM52XocGgGo3S-

Meetings Cookstown, Dublin & Cork in run up to EUFIC Regulation
Challenges & Developments

• What is food allergy?
• Why increasing?
• Is there a cure?
• Information for Consumers
• Enforcement science
• Interpretation?
• Supply chain security

• Definitions
• Research
• Oral immunotherapy
• Well underway
• Results always right?
• Thresholds
• Cases
What is food allergy?

A facet of food hypersensitivity
What is Food Hypersensitivity?

ADVERSE REACTIONS TO FOOD

PREDICTABLE REACTIONS
[e.g. to pathogens or toxins]

UNPREDICTABLE REACTIONS

REPRODUCIBLE

NON REPRODUCIBLE [e.g. food aversion]

FOOD HYPERSENSITIVITY

IMMUNE- MEDIATED
food allergy
IgE and non-IgE

OTHER MECHANISMS
[e.g. coeliac condition
& lactose intolerance]

Food hypersensitivity – objectively reproducible symptoms or signs initiated by a defined stimulus at a dose tolerated by ‘normal’ subjects*

*EAACI Position Statement: Johansson et al., 2001, Allergy 56:813

www.safefoodallergy.ning.com
• In the spectrum of adverse reactions to food there are predictable events – if anyone eats a sandwich containing a large number of pathogenic *Salmonella* they will be ill.

• Some reactions aren’t predictable; that is to say, until they happen, you don’t know they will happen, and not everyone is affected.

• When those reactions happen almost every time the person eats that food the reactions are ‘reproducible’.

• Reproducible adverse reactions to some foods are termed ‘food hypersensitivity’, objectively reproducible symptoms or signs initiated by a defined stimulus at a dose tolerated by ‘normal’ subjects, and which can take many forms.

• Broadly, if the hypersensitivity is mediated by your immune system it is food allergy, specifically an immune protein called immunoglobulin E, IgE.

• There are other mechanisms such as coeliac condition or more broadly food intolerance such as lactose intolerance.

• Its not as simple as this and we need to do a lot to find out more about what causes each type, and the grey areas in between ....
Sensitisation

The Nose
Aeroallergens

DENDRITIC CELL

ALLERGEN

GASTRO INTESTINAL MUCOSA / DAMAGED DERMAL EPITHELIUM

Naive T CELL

IL4

Th2

B CELL

IL4, IL13

ALLERGEN-SPECIFIC IgE

MAST CELL

After De Leon *et al.*

www.safefoodallergy.ning.com
**Challenge**

**ALLERGEN**

**GASTRO INTESTINAL MUCOSA**

**RELEASE OF**

**INFLAMMATORY MEDIATORS**

**GASTRO INTESTINAL SYMPTOMS**
- Mouth Swelling
- Diarrhoea
- Vomiting

**EXTRA-INTESTINAL SYMPTOMS**
- Pruritus
- Asthma
- Urticaria
- Angioedema
- Anaphylaxis

[www.safefoodallergy.ning.com](http://www.safefoodallergy.ning.com)
And sometimes – though rarely

Ulster schoolgirl dies after eating peanuts
Allergic reaction blamed for teen’s death

Belfast Telegraph 2006

Caterer Liable for Death of Wedding Guest - Egg Allergy Update
Jan 22 2010

A caterer who served a dessert containing egg that killed a man with a rare allergy at a Sikh wedding, has lost his appeal against a Court Order that he pay compensation to the widow.

Metro 2012

Emma Egerton died after just one mouthful of curry from an online takeaway that failed to provide a ‘may contain peanuts’ warning (Picture: Cavendish)
Fatal food anaphylaxis

Annual incidence rate for different events in food allergic people:

(a) Fatal food anaphylaxis

(b) Fatal food anaphylaxis in people aged 0-19

Fire / murder

Lightning

Clinical & Experimental Allergy
Volume 43, Issue 12, pages 1333-1341, 22 NOV 2013 DOI: 10.1111/cea.12211
What factors influence the severity of food allergic reactions?

Is it possible to identify thresholds of exposure to allergenic foods that are required for sensitisation and allergic reaction?

Why are some food proteins allergenic when others aren't?

Oral or dermal exposure?

Early exposure or avoidance?

There are many unanswered questions

Learning Early About Peanut Allergy

Integrated Approaches to Food Allergen and Allergy Risk Management (iFAAM)

www.safefoodallergy.ning.com
The major food allergens

- EU – 1169/2011 Annex II -- 14 substances or products
- Previously Annex III(a) of Directive 2000/13/EC (as amended)
  - 1. Cereals containing gluten, namely: wheat, rye, barley, oats, spelt, kamut or their hybridised strains, and products thereof,* wheat (such as spelt and khorasan wheat), rye, barley, oats or their hybridised strains, and products thereof except: [ingredients that do not contain sufficient allergen protein to elicit a reaction] - wheat/barley based glucose syrups, maltodextrins,& cereals used to make alcoholic distillates
  - 2. Crustaceans and products thereof;
  - 3. Eggs and products thereof;
  - 4. Fish and products thereof, except fish gelatine (carrier for vitamin or carotenoid preparations) & fish gelatine or isinglass (fining beer and wine);
  - 5. Peanuts and products thereof;
  - 6. Soybeans and products thereof, except fully refined soybean oil/fat, natural mixed tocopherols etc from soybean sources; phytosterols / esters and stanol ester from soybean sources;

* (EU) No 78/2014 of 22 November 2013
– 7. **Milk** and products thereof (including lactose), except whey used for making alcoholic distillates including ethyl alcohol of agricultural origin, lactitol;

– 8. **Nuts**, namely: **almonds**, **hazelnuts, walnuts, cashews**, **pecan nuts**, **Brazil nuts, pistachio nuts**, **macadamia or Queensland nuts**, and **nut products except alcoholic distillates** … …

– 9. **Celery** and products thereof;

– 10. **Mustard** and products thereof;

– 11. **Sesame seeds** and products thereof;

– 12. **Sulphur dioxide** and sulphites at concentrations of more than 10 mg/kg or 10 mg/litre in terms of the total SO₂ which are to be calculated for products as proposed ready for consumption or as reconstituted according to the instructions of the manufacturers;

(underlined text is the only difference between the old Directive and the new Regulation as regards SO₂)

– 13. **Lupin** and products thereof;

– 14. **Molluscs** and products thereof.

- Article 9 Mandatory particulars … include Annex II substances

- Article 21 … indicated in the list of ingredients with a clear reference to the name … emphasised through a typeset that clearly distinguishes it from the rest of the list of ingredients

- Article 44 … Includes non-prepacked, PPDS and catering sales,

- Art. 36 - The Commission shall adopt implementing acts on the application of requirements … [on] … voluntary food information:
  – (a) information on the possible and unintentional presence in food of substances or products causing allergies or intolerances
EUFIC

How well are we doing?
Food security for the food sensitive consumer on the island of Ireland

SafeFood funded – QUB UCC

- How food sensitive consumers make their food choices
- How they decide whether to trust allergen ingredient and precautionary labelling
- Any opportunity for industry to improve the range and quality of products available to food hypersensitive consumers
- Availability and cost of testing for food allergens, food intolerance parameters and gluten

Dr Tassos Koidis QUB
Dr Alexandros Stratakos QUB
Dr Audrey Dunn Galvin UCC
M Walker MChemA
H Gowland FIFST
M Bell FIFST
Catering Outlet Dublin Airport

Dear Customers,
If you would like to know more about food allergies, please see our booklet located at the counter.

All images © Hazel Gowland
Allergies

Before you order your food and drink, please speak to our colleagues if you have a food allergy or intolerance.

We can tell you all the specified allergens that are deliberate ingredients in our products.

As our kitchen handles foods containing flour, eggs, milk, nuts and other allergens, there is always a risk of cross contamination so we cannot guarantee that any product is entirely free from any allergens.
### Table

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Item 1</td>
<td>Description 1</td>
</tr>
<tr>
<td>Item 2</td>
<td>Description 2</td>
</tr>
<tr>
<td>Item 3</td>
<td>Description 3</td>
</tr>
<tr>
<td>Item 4</td>
<td>Description 4</td>
</tr>
</tbody>
</table>

All images © Hazel Gowland
Retail outlet
Dublin Airport

All images © Hazel Gowland
Retail outlet – counter sign

CUSTOMERS

Hot food must be consumed within 90 minutes of purchase or refrigerated and consumed within 24 hours.
Cafe Dublin City Centre
Laminated Allergen information
Cafe Dublin City Centre
Separate Gluten Free section

All images © Hazel Gowland
Dublin Window sign

All images © Hazel Gowland
Why is food allergy increasing?
Why the increase?

- Immunology - why do some people develop allergen-specific IgE antibodies to harmless food proteins and go on to exhibit food allergy, and why is this increasing in the developed world?
- Truthful answer is we don’t know…
- Fewer parasites
- Less diverse microbiome ... environmentally & personally
- Increasing new foods
- Air Pollution
- Genetics / epigenetics
- Weaning practices
- ……

Research – EAACI & the safefood Network will keep you posted
Gideon Lack, Department of Paediatric Allergy, St Thomas’ Hospital,

Fox, et al., 2009, J Allergy and Clin Immunol 123, 417-423;
Du Toit et al., 2008 J Allergy and Clin Immunol, 122, 984-991
Is there a cure?
Is there a cure? No – but oral immunotherapy, OIT, very promising Addenbrookes Study

- 99 peanut allergic children recruited to treatment and control groups
- Increasing amounts of peanut flour by mouth over 5 months
- 84% treated group tolerated 800 mg peanut protein (~5 peanuts)
- Control group retained their peanut allergy unchanged
- Children in control arm offered OIT, 91% could eat at least five peanuts a day at the end of the therapy

Assessing the efficacy of oral immunotherapy for the desensitisation of peanut allergy in children (STOP II): a phase 2 randomised controlled trial, Anagnostou et al., Lancet January 30th 2014
Food allergen protein analysis
What are the Options for Analysis?

- ELISA
- lateral flow devices designed for rapid on site testing
- DNA
- LCMS

[Images of ELISA, lateral flow device, DNA, LCMS]
## Main laboratory techniques

<table>
<thead>
<tr>
<th>Technique</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>ELISA</td>
<td>Routine, quantified results, good precision but results can be kit dependent and questionable recovery. Targets proteins (allergens?) therefore of direct relevance to allergic consumers. Relatively inexpensive if batched</td>
</tr>
<tr>
<td>PCR</td>
<td>Detects DNA not protein hence not probative of the presence of allergen but sensitive. DNA &gt; stable than proteins (harsher extraction conditions). Not quantitative</td>
</tr>
<tr>
<td>Real time PCR</td>
<td>As PCR but might be made quantitative.</td>
</tr>
<tr>
<td>Mass Spectrometry</td>
<td>Promise of bringing the gold standard for small molecule analysis to allergen proteins - molecular confirmation of identity, quantification, multiplex analysis and metrological traceability.</td>
</tr>
</tbody>
</table>


Enzymatic digestion

Peptides released from protein

Protein

Labelled peptide

When thresholds/action levels adopted – how do we analyse for allergens?
Liquid chromatography with tandem mass spectrometry
protein quantification

T5: HGLDNYR

T7: FESNFNTQATNR

T16: GTDVQAWIR

LC-MS/MS

www.safefoodallergy.ning.com
Different methods (ELISA kits) give different results

- Normalising spiked sample

Sykes, Anderson & Parmar, 2012, Normalisation of data from allergens proficiency tests, Anal & Bioanal Chem, 403, 3069

Global improvement in food allergen results ELISA, PCR, MS

Use of a well characterised material to aid quality control of results has been endorsed by many

e.g. Diaz-Amigo and Popping, J. AOAC Int, 2010, 93(2), 434-441
Traceable results relevant to food industry and allergic consumers

**Clinical Studies**

Food material designed to be used in a clinical setting presents difficulties but affords unrivalled opportunities for a material that links laboratory analysis to clinical effects
New – peanut allergen QC materials

- Two materials
  - negative control
  - positive control containing 10 mg/kg peanut protein added as light roasted peanut flour
- Based on EuroPrevall dessert mix
- Give a link with clinical studies
- Prepared by University of Manchester
- Sealed in nitrogen-flushed foil sachets containing 5 g dessert mix
- Prepared ≈ 900 sachets of each material
- Paste has to be reconstituted before use
Interpreting the results of analysis
-- ‘thresholds’
Thresholds - What do we mean by ‘threshold’?

- Limits for allergen proteins in food below which most of the food allergic population will not react
- Amount predicted to produce a reaction in a defined proportion of the population - protection of the vast majority & acknowledges impossibility of proving zero risk or absolute certainty

Crevel, et al., 2007, Food and Chem Toxicol, 45, 691-701,

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Fig. 1. Graphs showing the difference between an unbounded distribution (no threshold implied) and a bounded distribution, where a threshold of no response is implied.
But …the severity of allergic reaction depends on…

- Genetics
  - atopy, may be $10^6$ difference between least & most sensitive
- Dose
- Matrix – e.g. fat, pH, binding….
- Processing
- Exercise
- Medication (NSAIs)

- Alcohol
- Asthma
- Concurrent or recent infection
- Individual
  - Age, knowledge experience
- Situation
- .....
What would ‘thresholds’ look like and what do we need to know?

• Clinically derived individual thresholds – best practice the DBPCFC
• Transferred to a population basis using adequate sample size and good characterisation of the tested population wrt the whole allergic population
• Typical cross contamination concentrations in food

Crevel, et al., (2008), Thresholds for food allergens and their value to different stakeholders. Allergy, 63: 597–609

Crevel, et al., 2007, Food and Chem Toxicol, 45, 691-701,
The Allergen Bureau VITAL grid

Typical protein levels in allergenic foods and ACTION LEVELS with caveats
Allergen Bureau Action Levels

- Intentionally added allergens must be declared on the product label (e.g. in the L/I).
- Action Levels are the concentrations which define the labelling outcomes for each concentration of cross contact allergen. They are determined using the Reference Dose and the Reference Amount/Serving Size.
- Review cross contact allergens for opportunities to reduce or eliminate from the product.
- Where they cannot be eliminated, cross contact allergens should be labelled as specified by the appropriate Action Level:
  - **Action Level 1** – precautionary cross contact statement is not required for the relevant allergen under evaluation
  - **Action Level 2** – precautionary cross contact labelling statement is required for the relevant allergen using the standard VITAL statement.
- Precautionary labelling should only be used after a thorough assessment of the risk. Precautionary cross contact statements must **NEVER** be used as a substitute for good manufacturing practice (GMP) or as a generic disclaimer. Every attempt must be made to eliminate or minimise cross contact by adhering to GMP.
- The **ONLY** precautionary statement to be used in conjunction with VITAL is: “May be present: [name of allergen]”
VITAL Examples – egg and soya but you MUST read the whole document!

- AB recommends use their guide and excel spreadsheet (free…)
- Calculation is:-

$$\text{Action Level transition} = \frac{\text{Ref Dose (mg)}}{\text{Ref Amount (g)}} \times 1000$$

**Reference Dose** the milligram protein level (total protein from an allergenic food) below which only the most sensitive individuals (between 1% and 5% depending on the quality of the data set available) in the allergic population are likely to experience an adverse reaction.

**Reference amount** = defined by manufacturer and is the maximum amount of a food eaten in a typical eating occasion. This may be the same as the “serving size”
Example 1 Soya

- Reference dose soya = 1 milligram soya protein
- Reference amount (serving size) = 40 g

- Action Level transition point = 1*(1000/40) = 25 mg kg\(^{-1}\)
  - Above 25 mg soya protein per kg product precautionary cross contact labelling statement is required for the relevant allergen using the standard VITAL statement.
  - Can convert soya protein to soya using the protein content of the soy ingredient you are using or the AB typical levels

- If serving size is 100 g transition is 1*(1000/100) = 10 mg kg\(^{-1}\)
Example 2 Egg

- Reference dose egg = 0.03 milligrams egg protein
- Reference amount (serving size) = 40 g

- Action Level transition point = 0.03*(1000/40) = 0.75 mg kg\(^{-1}\)
  - Above 0.75 mg egg protein per kg product precautionary cross contact labelling statement is required for the relevant allergen using the standard VITAL statement.
  - Can convert egg protein to egg using the protein content of the egg ingredient you are using or the AB typical levels

- If serving size is 100 g transition is 0.03*(1000/100) = 0.3 mg kg\(^{-1}\)
Examples of ‘Thresholds’

<table>
<thead>
<tr>
<th>Protein Type</th>
<th>VITAL ED1-5 %</th>
<th>Defernez, … Mills et al ED 10 %*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg protein</td>
<td>0.03 mg</td>
<td>0.6 mg (0.1 – 2.3)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.3 mg (0.3 mg – 4.8)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.0 mg (0.2 – 3.6)</td>
</tr>
<tr>
<td>Milk protein</td>
<td>0.1 mg</td>
<td>0.1 mg (0.03 – 0.7)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.2 mg (0.03 – 1.1)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.1 mg (0.02 – 0.8)</td>
</tr>
<tr>
<td>Peanut protein</td>
<td>0.2 mg</td>
<td>2.8 mg (0.2 – 36)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>6.6 mg (0.5 – 51.7)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5.2 mg (0.4 – 41.9)</td>
</tr>
</tbody>
</table>


- Objective symptoms
- Mean and (95 % confidence interval)
- Lognormal, loglogistic, Weibull non-parametric curve fits and interval censoring
- Egg and milk are in the paediatric population (<3.5 years)
## Proposed ‘reference doses’

**EAACI Food Allergy & Anaphylaxis Guidelines**

<table>
<thead>
<tr>
<th>Food</th>
<th>Reference Dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peanut ED 1 %</td>
<td>0.2 mg peanut protein</td>
</tr>
<tr>
<td>Cow’s milk ED 1 %</td>
<td>0.1 mg milk protein</td>
</tr>
<tr>
<td>Egg ED 1 %</td>
<td>0.03 mg egg protein</td>
</tr>
<tr>
<td>Hazelnut ED 1 %</td>
<td>0.1 mg hazelnut protein</td>
</tr>
<tr>
<td>Soya ED 5 %</td>
<td>1.0 mg soya protein</td>
</tr>
<tr>
<td>Wheat ED 5 %</td>
<td>1.0 mg wheat protein</td>
</tr>
<tr>
<td>Cashew ED 5 %</td>
<td>2.0 mg cashew protein</td>
</tr>
<tr>
<td>Mustard ED 5 %</td>
<td>0.05 mg mustard protein</td>
</tr>
<tr>
<td>Lupin ED 5 %</td>
<td>4.0 mg lupin protein</td>
</tr>
<tr>
<td>Sesame seed ED 5 %</td>
<td>0.2 mg sesame protein</td>
</tr>
<tr>
<td>Shrimp ED 5 %</td>
<td>10 mg shrimp protein</td>
</tr>
<tr>
<td>Fish ED 5 % (provisional)</td>
<td>0.1 mg fish protein</td>
</tr>
</tbody>
</table>

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*Muraro et al., Allergy, J Allergy Clin Immunol DOI: 10.1111/all.12453*

*Taylor et al., 2014, Food Chem Toxicol, 63, 9 – 17*

*Allen et al., 2014, J Allergy Clin Immunol, 133, 156 – 164*

*Alvarez & Boye, 2012, J Allergy (Cairo), 2012:746125*

**EFSA consultation on a draft scientific opinion on the evaluation of allergenic foods and food ingredients for labelling purposes**
Supply chain security
Secure supply chain?

• February 2013

Food fraud: Deadly peanuts may be lurking in your high street takeaway

UNDERCOVER investigators prevented a peanut allergy disaster when they exposed a high street food fraud.
Food crime

• TSOs Cumbria bought chicken tikka masala from two restaurants & were assured peanuts had not been used.
• Public Analyst found peanut protein rather than almond.
• Ingredients were traced back to one of the country’s biggest Indian food suppliers.
• Euro Foods, was fined £6,000 with £12,000 costs after being found guilty of food adulteration but successfully appealed the conviction – summons was ‘bad for duplicity’...

Cumbria TSO “Peanuts are a cheaper food than almonds and it is absolutely unacceptable that the almonds were substituted and sent to the takeaway outlets, which are not at fault. In the worst-case scenario, the actions of Euro Foods could have been life-threatening for some people”
.... bad for duplicity...

- Eurofoods grounds of appeal - error in law of laying information (charges) under both sections 15(2) and 15(3) of the Food Safety Act 1990 (Rule 12 MCR)

- Offences:
  - S15(2) falsely describing or misleading as to nature or substance or quality
  - S15(3) presentation of which is likely to mislead as to the nature or substance or quality

- Rule 12 MCR preserved in the Criminal Procedure Rules 2005,

- But from 2008 seems to be permissible for an information to charge more than one offence so long as those offences are described within separate allegations.


How long has this been going on?

Woman is killed by two spoons of a curry

Daily Mail Reporter

A young woman with a severe nut allergy died after eating just two spoonfuls of a takeaway curry.

Kate Obertelli had forgotten the life-saving drug she usually carried with her in case of such emergencies.

The adrenaline injection pen lay ten miles away from her home.

The 31-year-old is believed to have warned staff at the takeaway of her allergy when placing her order.

But yesterday her family refused to start a ‘witch hunt’ against the unnamed restaurant.

‘There has been a mistake made but we cannot be angry about that,’ said her heartbroken father Mike, who described Miss Obertelli as a ‘lovely, bubbly’ girl.

‘I lost my only daughter and she was the love of my life.

‘Now you just have to concentrate on’

Emma Egeton died after just one mouthful of curry from an online takeaway that failed to provide a ‘may contain nuts’ warning (Picture: Cavendish)
Consequences

• Anaphylaxis Campaign has collected details of fatal and near-fatal reactions since 1992
• 7 - 10 food allergy deaths a year, mostly from peanuts but also tree nuts, milk, fish, shellfish and egg.
• Peanut allergy is the most common.
• The campaign’s food adviser Hazel Gowland said:
  • “This case confirms what has long been suspected, that some ingredients in the supply chain for curry takeaways are not what they seem and may explain some of the severe food allergic reactions reported around the UK.”
  • "At least one fatal reaction per year as well as a number of ‘near-misses’ involve a curry or similar takeaway meal”
FSA investigations

• During 2012 FSA investigated severe allergic reactions following the consumption of curry dishes purchased from Indian restaurants and take-aways. Some of these incidents resulted in fatalities.
• Some of these incidents were caused by the use of a ground almond ingredient, which also contained ground peanut.
• FSA identified
  – “weaknesses in the food chain where the contamination and loss of clear information occurred”
  – Poor understanding of the significance of substituting peanuts for almonds and incorrect allergen information provided at a point of sale
  – unclear labelling and confusion between peanuts and tree nuts (almonds) leading to the potential for accidental substitution.
  – possible economically motivated adulteration, driven by the financial incentive to substitute ground almonds with ground peanut.

FSA Annual Report of Incidents 2012
What needs done?

• FSA
  – Issued a web story, provided risk assessments to local authorities, encouraging local authorities to identify underlying trends, continue to work with catering trade associations to raise awareness

• Is this enough?
• Connor Donaldson died Oct 2013 from an anaphylactic reaction to peanuts, inquest 24th March 2014 (Leigh Journal).
• Police ruled out criminal behaviour but EHO investigation ongoing
• EHO said: “On a routine inspection at the takeaway, we found staff had a poor knowledge of allergies ….
• Added there was another potential threat of cross contamination as an almond powder bought in by the takeaway and used in dishes such as Kormas, was found to contain 50 per cent peanuts.
Man almost dies from takeaway 10 years after sister died the same way

The Bolton News

- Sale of nut free chicken tikka masala and nut free plain naan bread, customer with a known allergy to peanut, and who had specifically requested a nut free meal.

- Two spoonfuls of the meal .... severe anaphylactic shock and was admitted to hospital.

- 340 mg kg\(^{-1}\) of peanut protein in meal

First offence: s 14 Food Safety Act 1990 (the chicken tikka masala) – Fine £500; Second offence: s 14 Food Safety Act 1990 (the naan bread) – no separate penalty

Compensation to Mr Obertelli - £2,500.00

Costs £5,000.00

Subject to any appeal against sentence.
And then along came the cumin and paprika recalls ...
Cumin (Cummin)

An umbelliferous fruit + anise, caraway, Celery, coriander, dill & fennel. Latin *umbella* = parasol, sunshade. *Cuminum cyminum* family Apiaceae (celery, carrot or parsley family)

**Essential oil - 2.3 to 5%, of which 40–65% is cuminaldehyde.**

![Cumin Seeds](http://www.savoryspiceshop.com/content/mercury_modules/cart/items/2/7/8/2783/cumin-seeds-whole-organic-1.jpg)

![Umbelliferous Plant](http://www.paulnoll.com/Oregon/Wildflower/Flora/flower-inflorescence-choices-04.html)

![Chemical Structure](http://www.savoryspiceshop.com/content/mercury_modules/cart/items/2/7/8/2783/cumin-seeds-whole-organic-1.jpg)

| Carbohydrate | 44 % |
| Lipids       | 22 % |
| Protein      | 18 % |

*Cuminum cyminum - Köhler’s Medizinal-Pflanzen-198”* by Franz Eugen Köhler, Köhler’s Medizinal-Pflanzen
Cumin standards & adulteration

<table>
<thead>
<tr>
<th>Specification</th>
<th>Suggested limit</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>ASTA cleanliness specifications</em></td>
<td></td>
</tr>
<tr>
<td>Whole insects, dead (No.)</td>
<td>4</td>
</tr>
<tr>
<td>Mammalian excreta (mg/lb)</td>
<td>3</td>
</tr>
<tr>
<td>Other excreta (mg/lb)</td>
<td>5</td>
</tr>
<tr>
<td>Mould (% by weight)</td>
<td>1</td>
</tr>
<tr>
<td>Insect-defiled/infested (% by weight)</td>
<td>1</td>
</tr>
<tr>
<td>Extraneous foreign matter (% by weight)</td>
<td>0.5</td>
</tr>
<tr>
<td>Ash (% max)</td>
<td>9.5</td>
</tr>
<tr>
<td>Acid-insoluble ash (% max)</td>
<td>1.5</td>
</tr>
<tr>
<td><em>FDA DAL</em></td>
<td></td>
</tr>
<tr>
<td>Volatile oil (% min)</td>
<td>2.5</td>
</tr>
<tr>
<td>Moisture (% max)</td>
<td>9.0</td>
</tr>
<tr>
<td>Ash (% max)</td>
<td>8.0</td>
</tr>
<tr>
<td>Acid-insoluble ash (% max)</td>
<td>1.0</td>
</tr>
<tr>
<td>Average bulk index (mg/100g)</td>
<td>240</td>
</tr>
<tr>
<td><em>USFDA DAL</em></td>
<td></td>
</tr>
<tr>
<td>Sand and grit (AOAC 975.48)</td>
<td>Average of 9.5% or more ash and/or 1.5% or more acid-insoluble ash</td>
</tr>
</tbody>
</table>

**Adulterants**
- Caraway
- Sand, grit & debris
- Milled foliage, straw, wheat & rice husks
- Starches
- Olive stones
- Essential oil can be adulterated with synthetic cumin aldehyde
- Peanut & Prunus *spp* protein

Current episode of cumin adulteration

• At the outset not clear whether its contamination or deliberate adulteration
• Oct 2014 Canadian Food Inspection Agency random tests for allergens revealed undeclared peanut & almond protein in products containing cumin
• Widespread recalls of cumin products in N America (42 to 19 Feb 2015)
  – one for peanut and almond starting in Autumn 2014, and a second larger one for peanut only, starting in December 2014
  – Salsas, spices inc paprika & mixes, seasonings → meat products, hummus,
  – None of the N American recalled products distributed in UK

Seasoning and Spice Association Q&A
More ..... + UK & Europe recalls

- FDA reports ~dozen allergic reactions, severity of them is not clear
- Traced to two separate Turkish suppliers, no evidence were related
- Jan 31 FSA recalled ground cumin with almond protein, precautionary
- February 12th and 14th FSA issued two further recalls re undeclared almond protein in fajita meal/dinner kits and seasoning mixes
- Appears that a batch of paprika was the likely source (Santa Maria)
- Denmark, Sweden & Norway have issued alerts / recalls
- None of the tests have detected peanut proteins at levels that would require allergen labelling (FSA)
- Source was initially though might be
  - Cross contamination in harvesting, transport, storage, processing...
  - Peanut hulls, almond shells
  - Peanut meal (animal feed after oil extraction ...
Undeclared allergens RASFFs
1.1.15 – 1.3.15

24/02/15 GB Peanut 41.3 mg/kg milk chocolate biscuits from UK
17/02/15 GB Almond >18 mg/kg spice mix from Sweden, via UK
14/02/15 GB Almond >18 mg/kg fajita dinner kit from Sweden
13/02/15 GB Almond 306 mg/kg ground cumin from Turkey
12/02/15 GB Almond 270 mg/kg fajita meal from Sweden
12/02/15 IT Gluten in gluten-free milk chocolate bar from Italy
30/01/15 IT Milk ingredient and celery in meat sauce from Italy
28/01/15 DK Hazelnut and almond in peanut butter from the Netherlands
20/01/15 DK Wheat in noodles from Thailand
13/01/15 SK Soya >100 mg/kg candies from Hungary
But ... Mahleb ...

- Barts Ingredients Company Ltd, the manufacturer of the UK cumin recalled have claimed that mahleb, a little known nut, was possibly the origin of the ELISA almond positives
- 30 April 2015 CFIA rescinded ~ 25 product recalls ...
  - “... new evidence regarding the cross-reactivity of mahaleb, a spice obtained from a specific species (*Prunus mahaleb*) of cherry seeds, with the almond allergen test kit. It is highly likely that the positive sample results for the ground cumin and cumin-containing products were due to mahaleb contamination and not almond”
- Had been working on LC-MS/MS for peanut, applied method to almond and mahleb...
- Mahaleb (also mahlab, white mahlab ,mahleb, English cherry, Rock cherry, St. Lucie cherry (*Prunus mahaleb* L. of the Rosaceae family, subfamily Prunoidae)
Mahaleb (also mahlab, mahlab, mahleb, English cherry, etc... 

(*Prunus mahaleb* L.))

- Grown, for example in Turkey, where the flesh and seed of mahlab fruit are important.

- Mahlab puree, mahlab vine, mahlab flour and mahlab oil are produced from mahlab fruit. The mahlab seeds form an important source of protein and oil.

- The stones are cracked to extract the seed kernel, which is about 5 mm diameter ...

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Analysis for almond and other Prunus spp

- ELISA... Screening

- DNA – bioinformatics deficit... But developing real time PCR assays and sequencing ... 

- LC-MS/MS unique peptides identified
Standards - no authoritative allergen reference dose for almond, but ...

- Ref doses 0.2 milligrams peanut protein and 0.1 milligrams hazelnut protein (Muraro et al.),
- Assume almond allergic individuals react to as little as 0.1 mg almond protein
- 50 gram portion, 0.1 mg almond protein is a concentration of 2 mg/kg
- Almond ~ 20 % protein 2 ppm almond protein is 10 ppm whole almond.
- ELISA can detect almond at 10 ppm with little difficulty, and PCR is likely to be more sensitive.
- Testing is thus viable but must be validated in the matrix. There are as yet no reference or quality control materials for almond to help labs validate.
- Cumin and other spices used at low levels (say around 1 %) in most foods higher concentrations than 10 ppm of almond in the cumin would be required to trigger an allergic reaction in most people eating the finished food ... 1000 ppm?
- BUT - varying concentrations across the cumin batch, poor recovery, MU, extra sensitivity on the part of the allergic person, all ⇒ need for a safety margin so that 10 ppm almond in cumin is a good starting point for a limit of detection and lower if possible.
The future ...
• Research on causes & ‘cures’ will yield good useable results, but need much more work on fundamental immunology e.g. Treg ... and nutrition ...
• Food businesses hold responsibility for dealing with allergen cross contamination – thresholds will help but needs >> dialogue & work
• Thorough investigation of food allergy deaths, particularly in the catering sector, is required; the skills & capacity should be improved
• Knowledge & skills gaps re investigation & prosecution of potentially serious incidents of food allergen mismanagement and mislabelling
• Tenacious approach required - e.g. early realisation that samples of food and/or stomach contents should be retained and analysed
• Analysis urgently needs to be improved... More reference materials, better bioinformatics ... Metrological traceability needed ...
• Supply chain of the meal ingredients must also be rigorously followed up to find out where adulteration or contamination with the fatal allergen occurred
• EUFIC – good progress but robust enforcement on non-compliers needs to start soon
Food allergy – a forensic perspective

Michael Walker and Hazel Gowland discuss the latest allergen measurement techniques and review allergen non-compliance cases in the UK courts.

Review

Food allergy, a summary of eight cases in the UK criminal and civil courts: effective last resort for vulnerable consumers?

M. Hazel Gowland and Michael J Walker

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Thank you

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www.safefoodallergy.ning.com