Food Allergen Detection and Control Research at IFSH

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IFSH 2019 Annual Meeting

September 25, 2019
Topics

• Background
• Challenges with Food Allergens
• Allergen Research Focus at IFSH

• Collaborative Projects:
  o Retail Project:
    o Wiping, Cleaning and Transfer
  o Allergen Transfer in Shared Cooking Oil:
    o Shrimp, Breaded Shrimp, French Fries
  o Chocolate Pilot Plant Studies:
    o IFSH and Penn State University (PSU)
Major Allergens in U.S.

- Peanuts
- Tree Nuts
- Milk
- Eggs
- Soy
- Wheat
- Fish
- Crustacean Shellfish
Major Public Health Concern

- Food allergies reported: up to ~ 4% adults and ~8% children
- Nearly 40% of food allergic children have multiple food allergies
- Center for Disease Control (CDC) study in 2013 noted ~ 50% increase in food allergy prevalence in children from 1997-2011
- More than 200,000 ambulatory-care visits/year to hospitals
- Hospital discharges reached ~9,500 children in 2004-2006
- Strict avoidance; still no cure

Gupta et al. Pediatrics, 2018; 142(6)
Gupta et al. JAMA Network Open, 2019
Allergen Regulations in US

Food Allergen Labeling and Consumer Protection Act (FALCPA), 2004

• Declare major food allergen ingredients on label
• Cross-contact not covered by FALCPA
• Advisory or precautionary labels are voluntary and not regulated
  o “may contain”, “processed in a facility …”

Food Safety Modernization Act (FSMA), 2011

• Prevent allergen cross-contact
• Control of allergens: label, cross-contact, sanitation, supplier controls
Cross-Contact: How Does it Occur?

- Dust or aerosols containing allergens
- Improper use of rework
- Cross-over points in processing lines
- Employee errors
- Reuse of cooking oil
- Ineffective cleaning
- Other
Some Challenges

Industry:
- Proper labeling
- Adequate cleaning
- Cross-contact
- Supply controls
- No regulatory limit

Consumer:
- Risk of allergic reaction
- Advisory/precautionary statements

FDA:
- Analytical challenges
  - Processing effects
  - Food matrix
  - Reference materials
- No regulatory limit
Allergen Research Goals at IFSH

• Trace the fate and detectability of allergens/gluten

• Evaluate effectiveness of various cleaning and other control strategies

• Investigate allergen transfer
  o Simulate allergen cross-contact: lab and pilot plant
Three Past Collaborative Projects

- **Research Challenges**
  - Trace the fate and detectability of food allergens/gluten

- **Projects**
  1) Pilot scale production of cereal bars/muffins and impact of thermal treatment on allergen quantitation *(General Mills)*
  2) Detection of soy/gluten in soy sauce during fermentation *(Kikkoman)*
  3) Gluten detection in barley based beer with/without prolyl endopeptidase enzyme *(pilot plant: University of Wisconsin-Madison)*

- **Outcomes**
  - Developed complimentary analytical methods to ELISA *(LC-MS/MS)*
  - Evaluated allergen cross-contact and cleaning methods
  - Determined effects of processing on detectability
Detection of gluten in a pilot-scale barley-based beer produced with and without a prolyl endopeptidase enzyme

Katherine L. Fiedler, Wanying Cao, Liyun Zhang, Magdalena Naziemiec, Binaifer Bedford, Lanlan Yin, Nicholas Smith, Matthew Arbuckle, Arnoldo Lopez-Hernandez and Lauren S. Jackson

1) J Agric Food Chem. 2015 Dec 16;63(49):10669-10680

2) Food Addit Contam Part A. 2019 Aug;36(8):1151-1162
Current Allergen Projects

• **Allergen removal/transfer in retail/food service environments**
  o FDA/CFSAN- OFS (Retail Food Protection Team)

• **Transfer of allergens in fryers using shared cooking oil**
  o IIT; FDA/CFSAN- OFS (Seafood Processing and Technology Branch), ORS

• **Milk cross-contact to dark chocolate on shared lines**
  o Penn State University; IIT; FDA/CFSAN- OARSA

• **Research Focus:**
  o Determine extent of cross contact in lab or pilot plant studies
  o Evaluate cleaning and other allergen control strategies
  o Investigate effects of heat and processing

• **Limited published research available**
Allergen Removal and Transfer in Retail/Food Service Environments

FDA Collaborators:
OFS: Retail Food Protection Team
Relevance/Impact: Allergen Retail Project

- **FDA Food Code**: Best advice on safety/protection of food for retail/food service establishments
- Provisions focus on microbial contamination/control
- Limited allergen references in FDA Food Code

**Project Goals:**
- Investigate **wiping** and **cleaning** methods for allergen removal
- Evaluate extent of allergen **transfer**
Wiping Study

Surfaces with no allergen → Allergen applied to surface → Surface wiped for 5 sec (one wipe/surface): → Determined presence of allergen with LFD test
Full Cleaning: Wash-Rinse-Sanitize-Air Dry Method

- Wash pail with detergent
- Rinse pail with tap water
- Sanitizer solution (50 ppm total chlorine)
- Air dry surfaces
Allergen Transfer Study

Mayonnaise (2 g) on wet cloth soaked in sanitizer solution

Surfaces after transfer study
Highlights

• Key Factors Affecting Allergen Removal:
  o Nature and amount of allergen; food contact surface; type and state of wipe/cloth

• Full Cleaning Method and Transfer Studies:
  o Wash-rinse-sanitize-air dry was effective, overall
  o Pre-scrape step improved removal on textured plastic
  o Use limitations with cloth storage in sanitizer, minimized transfer

• FDA Food Code (Chapters 3 and 4) effective in general, for allergen removal / minimize transfer
Allergen Transfer in Shared Frying Oils

Collaborators:
FDA/OFS: Seafood Processing and Technology Branch
FDA/ORS and IIT
Cross-Contact in Shared Fryers

**Batch/vat fryer**  
(home, restaurant, food industry)

**Continuous fryer**  
(food industry)

1) Shellfish, fin fish or battered/breaded/coated product → **Shared frying oil**

2) French fries (secondary food) → **Shared frying oil**  → **Allergen transfer**?

**Allergens:** Shellfish, fish; wheat, milk, egg…(breading/batter/coatings)
Relevance/Impact: Shared Fryers/Oil

Research Questions:
- What is the extent of allergen transfer and cross-contact that occurs with shared fryers and oil?
- What is allergen contribution from breaded/battered/coated products?
- What role does oil filtration play in allergen removal?

Provide FDA, National Marine Fisheries Service, retail and seafood industry information on allergen risk due to cross-contact in shared fryers.

Project Goal:
- Trace seafood allergens, gluten, milk → shared oil → secondary food
Shrimp Transfer to Oil and French Fries

Ten (100 g) batches of shrimp fried and four (100 g) batches of French fries
## Shrimp Transfer Study

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total frying time (min)</th>
<th>Maruha Crustacean protein ppm (%CV); LOQ: 0.78 ppb</th>
<th>ELISA System Tropomyosin levels ppm (%CV); LOQ: 0.05 ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unheated Control oil</td>
<td></td>
<td>&lt;LOQ</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Heated Control oil</td>
<td>0 min</td>
<td>0.14 ppm (1%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Oil after frying batch 2 shrimp</td>
<td>8 min</td>
<td>22.9 ppm (9%)</td>
<td>1.18 ppm (15%)</td>
</tr>
<tr>
<td>Oil after frying batch 4 shrimp</td>
<td>16 min</td>
<td>31.6 ppm (13%)</td>
<td>1.16 ppm (24%)</td>
</tr>
<tr>
<td>Oil after frying batch 6 shrimp</td>
<td>24 min</td>
<td>30.9 ppm (14%)</td>
<td>1.09 ppm (7%)</td>
</tr>
<tr>
<td>Oil after frying batch 8 shrimp</td>
<td>32 min</td>
<td>36.0 ppm (13%)</td>
<td>1.29 ppm (24%)</td>
</tr>
<tr>
<td>Oil after frying batch 10 shrimp</td>
<td>40 min</td>
<td>34.3 ppm (28%)</td>
<td>0.99 ppm (26%)</td>
</tr>
<tr>
<td>Oil after frying batch 1 French fries</td>
<td>44 min</td>
<td>3.18 ppm (12%)</td>
<td>0.07 ppm (21%)</td>
</tr>
<tr>
<td>Oil after frying batch 2 French fries</td>
<td>48 min</td>
<td>1.51 ppm (60%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Oil after frying batch 3 French fries</td>
<td>52 min</td>
<td>0.45 ppm (34%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Oil after frying batch 4 French fries</td>
<td>56 min</td>
<td>0.58 ppm (65%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Raw French Fries (Control)</td>
<td>-</td>
<td>&lt;LOQ</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Batch 1 fried French Fries</td>
<td>44 min</td>
<td>10.4 ppm (14%)</td>
<td>0.15 ppm (12%)</td>
</tr>
<tr>
<td>Batch 2 fried French Fries</td>
<td>48 min</td>
<td>6.27 ppm (85%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Batch 3 fried French Fries</td>
<td>52 min</td>
<td>1.61 ppm (21%)</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>Batch 4 fried French Fries</td>
<td>56 min</td>
<td>2.68 ppm (22%)</td>
<td>&lt;LOQ</td>
</tr>
</tbody>
</table>
Research Questions

- Measured level of shrimp protein relatively constant in oil, despite increased allergen load

  Why?

  Do we have the full picture?

- Published information lacking on cross-contact in oil

- Additional experiment: Frying time study on shrimp detectability
## Effect of Frying on Shrimp Allergen Detection

<table>
<thead>
<tr>
<th>Shrimp sample</th>
<th>Maruha Crustacean Protein ppm (%CV)</th>
<th>ELISA Systems Tropomyosin levels ppm (%CV)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw shrimp</td>
<td>686,000 ppm (6%)</td>
<td>14,600 ppm (18%)</td>
</tr>
<tr>
<td>Shrimp fried for 10 min</td>
<td>223,000 ppm (11%)</td>
<td>16,900 ppm (14%)</td>
</tr>
<tr>
<td>Shrimp fried for 20 min</td>
<td>106,000 ppm (10%)</td>
<td>6,150 ppm (7%)</td>
</tr>
<tr>
<td>Shrimp fried for 30 min</td>
<td>33,500 ppm (17%)</td>
<td>1,710 ppm (14%)</td>
</tr>
<tr>
<td>Shrimp fried for 40 min</td>
<td>22,400 ppm (88%)</td>
<td>683 ppm (30%)</td>
</tr>
<tr>
<td>Shrimp fried for 50 min</td>
<td>18,800 ppm (16%)</td>
<td>321 ppm (13%)</td>
</tr>
<tr>
<td>Shrimp fried for 60 min</td>
<td>5,390 ppm (14%)</td>
<td>137 ppm (8%)</td>
</tr>
<tr>
<td>Shrimp fried for 70 min</td>
<td>2,600 ppm (16%)</td>
<td>11 ppm (20%)</td>
</tr>
</tbody>
</table>

Allergen detectability increased as frying time increased.
Highlights and Challenges

- Shrimp protein **transfer detected** in oil and fries

- **Frying/heat** dramatically **decreased ability to detect shrimp proteins** in fried shrimp

- **Analytical challenges exist** due to heating allergen and extraction from oil

- **Need complimentary analytical methods to ELISA:** FDA/CFSAN/ORS
In Progress and Future Experiments:

**Allergens:**
- Tropomyosin
- Crustacean Protein
- Wheat
- Milk

**Breaded Shrimp:** Transfer of multiple allergens (in progress)

**Future work:**
- Continuous fryer experiments
- Filtration studies regarding re-use of oil
Effectiveness of Dry Cleaning Methods for Removing Milk Chocolate from Processing Lines/Valves and Equipment

- IFSH Pilot Plant Studies
- Penn State University (PSU) Pilot Plant Experiments

Collaborators: IIT, PSU and FDA/OARSA
Background

• Chocolate/Confections Category: High risk for milk-allergic consumers

  WHY?

• Shared processing lines
• Cross-contact with milk allergen
• Dry cleaning challenges

References:
Relevance/Impact

- **Simulate allergen cross-contact:**
  - Determine extent of milk carryover: milk chocolate to dark chocolate
  - Use shared processing line (pipe and valve only): IFSH Trial
  - Shared processing equipment: Penn State Trial

- **Evaluate effectiveness of various dry cleaning methods:**
  - No cleaning: push-through with dark chocolate only
  - Mechanical method (pig) followed by dark chocolate (IFSH Trial)
  - Cocoa butter flush and dark chocolate push-through
Some Dry Cleaning Strategies in Chocolate Processing

- Manual scraping
- Dark chocolate push-through
- Pig-purging of lines
- Cocoa butter flush

• Dry cleaning has limitations:
  o food residue may remain on line/equipment
  o allergen may carry-over from one product to another
Some Allergen Control Challenges in Chocolate Production

• Wet cleaning not typically used for some equipment
  o Microbial hazard, product incompatibility, equipment incompatibility

• Some types of equipment are difficult to disassemble/inspect

• Consistency of chocolate makes it difficult to remove from equipment

• Type/form of allergens in chocolate → different challenges for control (nut/peanut particulates vs. milk)

• Cross contact concerns
Pilot Plant Studies

IFSH Trials
Piping/valves
- Pipe + butterfly valve
- Pipe + ball valve

Penn State Trials
Chocolate manufacturing equipment
- Conche
- Ball Mill
- 3-Roll Refiner
Evaluation of Cleaning Procedures: Pipe/Valve

Schematic diagram of dark chocolate push-through tank with milk chocolate-contaminated pipe/butterfly or ball valve
## Results: Pipe/Ball Valve

<table>
<thead>
<tr>
<th>Dry cleaning method</th>
<th>Milk levels in initial samples ppm milk (%CV)</th>
<th>Milk levels in second samples ppm milk (%CV)</th>
<th>Kilograms of dark chocolate push-through to obtain &lt;LOQ</th>
</tr>
</thead>
<tbody>
<tr>
<td>No cleaning (pipe + valve)</td>
<td>11,000 (3.0% CV) – 15,000 (0.3% CV)</td>
<td>1,600 (3.4% CV) – 2,600 (7.4% CV)</td>
<td>20 – 22</td>
</tr>
<tr>
<td>Pig purging (pipe + valve)</td>
<td>&lt; LOQ</td>
<td>&lt; LOQ</td>
<td>NA</td>
</tr>
</tbody>
</table>

Samples were collected at 7-sec interval  
Flow rate of dark chocolate: ~ 147 g/sec  
CV: Coefficient of variation
Penn State Pilot-Scale Chocolate Processing Equipment

**Ball Mill** (max capacity ~500 g)
- Carbon steel balls
- Shaft

**Conche** (max capacity 5 kg)
- Vertically oriented shaft

**Refiner** (Lehman 3-roll refiner)
Chocolate in Conche

After draining dark chocolate

Cocoa butter flush for 5 minutes after draining milk chocolate
Results: Ball Mill and Conche with or without Cocoa Butter Flush

**Ball Mill**
- > 90% reduction

**Conche**
- > 88% reduction
Highlights

- **Pipe/valve assembly study (at IFSH)**
  - Cross-contact during changeover: milk → dark chocolate
  - Pig purging (most effective) < cocoa butter flush < no cleaning

- **Chocolate processing equipment study (at PSU)**
  - Cocoa butter flush on ball mill/conche dramatically decreased the level of milk found in dark chocolate
  - Milk cross-contact was observed in all experiments, regardless of the dry cleaning treatment used
  - Wet cleaning was effective in removing milk chocolate from ball mill and conche, but resulted in a long downtime
Visit Liyun Zhang’s chocolate poster for details

Effectiveness of cleaning strategies for removing milk chocolate from pilot-scale chocolate processing equipment
2018-2019 Allergen Team at Moffett

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IIT Staff/MS Student: Josh Warren
Acknowledgements

Lauren Jackson (FDA/IFSH)
Girvin Liggans (FDA/OFS)
Laurie Williams (FDA/OFS)
Nate Anderson (FDA/IFSH)
Girdhari Sharma (FDA/OARSA)
Quincy Suehr (FDA/IFSH)

Anirudh Kaja (IIT/IFSH)
Josh Warren (IIT/IFSH)
Liyun Zhang (IIT/IFSH)
Sakshi Gandhi (IIT/IFSH)

Allison Brown (PSU)
Helene Hopfer (PSU)
Greg Ziegler (PSU)

Anne Eischeid (FDA/ORS)
Steven Bloodgood (FDA/OFS)
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Karen Swajian (FDA/OFS)
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FDA- Office of the Chief Scientist
Thank You

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