Comparison of Visual Inspection, an Allergen-Specific Method (ELISA) and Nonspecific Methods (Sensitive ATP and Total Protein) to Detect the Presence of Allergenic Food Residues on Food-Contact Surfaces

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Abstract

Recent FDA and IFT surveys found that the most common method used by food manufacturers to check the effectiveness of their equipment cleaning program is visual inspection, followed by ELISA, bioluminescence/ATP, and other tests. The purpose of this study was to compare visual inspection, ELISA, ATP sensitive swabs, and total protein swabs for detecting the presence of allergenic food residues on food-contact surfaces. One ml of milk, egg, and peanut solutions (0-1000 µg/mL) were pipetted onto the surface of stainless steel, urethane, and Teflon plates. The plates were dried in an oven (80ºC) to obtain 0-1000 µg of the food residues. The plates were visually examined and then simultaneously swabbed for ELISA, sensitive ATP, and total protein testing.

In general, the detection limits (DL) for visual inspection were consistent with those determined by ELISA. The DL (visual, ELISA and ATP) for egg was 500 µg on all food contact surfaces, but lower with the total protein swabs. The ELISA and visual detection limit for peanut was 500 µg on all food surfaces, but 50-100 µg when tested with the ATP and total protein swabs. ELISA and visual tests detected milk at 100 µg on all food surfaces, but 10-50 µg when tested by sensitive ATP swab and total protein swabs. In some cases, total protein swabs detected food residues at or below levels detected by sensitive ATP swabs. This study shows that limitations for these methods exist and care must be taken when choosing a method for detecting the presence of food residues on food-contact surfaces.

Introduction

A recent IFT survey found that visual inspection is the method most commonly used for cleaning verification, followed by ELISA, bioluminescence/ATP, and other tests in all size and food product categories (Taylor et al., 2006). A recent FDA report found that the most common verification method is visual examination (95% of facilities), chemical assays for the allergen (5%), and other tests (6%) (USFDA, 2006).

A simple verification method for cleaning is visual inspection of equipment. Visual examination can be done only on equipment where food-contact surfaces and areas of potential food product and ingredient accumulation are readily accessible for inspection to enable observation of food soils, if present. There is a general assumption that the presence of visible residue indicates that allergens are present.

Verification of cleanliness is often performed using analytical methods. Immunoassays, such as ELISAs, are able to detect the presence of at least five different allergenic foods (egg, milk, peanut, soy, some tree nuts) on swabbed equipment surfaces or in rinse water. In general, the presence of food allergen in the finished product and in-process materials indicate a failure in the design or execution of the allergen cleaning protocol.

ATP (adenosine triphosphate) and total protein tests are also being used by the food industry to verify cleaning program effectiveness. These tests are rapid and inexpensive when compared to ELISAs, but do not directly measure the presence of food allergens. The ATP tests detect microbial ATP as well as ATP associated with residual foods and therefore, only can be used to verify the effectiveness of wet cleaning procedures. The total protein assays detect allergenic and nonallergenic proteins on food-contact surfaces and in solution. Research is needed to compare visual inspection to immunochemical (allergen-specific) methods and non-specific methods (ATP and total protein) for determining cleaning.

Objective

To compare visual inspection, ELISA, sensitive ATP, and total protein swabs for detecting the presence of allergenic food residues (milk, egg, and peanut) on food-contact surfaces.

Materials and Methods

ZCS whole milk powder (Charm Sciences, Inc., Lawrence, MA), egg powder (Honeyville Grain, Inc., Rancho Cucamonga, CA), and peanut flour (Kellogg's, Battle Creek, MI) were diluted with HPLC-grade water to obtain solutions of 0-1000 µg/ml. One ml of milk, egg, and peanut solutions (0-1000 µg/ml) were pipetted onto the surface of stainless steel, urethane-faced belting, and Teflon plates. The plates were dried in an oven (80℃) to obtain 0-1000 µg of the food residues. The plates were visually examined and then simultaneously swabbed for ELISA (Alert for peanut, egg, and total milk, Neogen Co., Lansing, MI) sensitive ATP (AllerGiene®, Charm Sciences, Inc., Lawrence, MA) and total protein (Aller-tect™, TECRA International, Australia) testing on all three food-contact surfaces for all three food allergens. All trials were performed at least three times.

Results

Table 1. Comparison of Detection Methods for Milk Residue on Food-Contact Surfaces

Surface	Method of Detection	Amount of Non-Fat Dry Milk Spotted on Surface					
		0 µg (control)	10 µg	50 µg	100 µg	500 µg	1000 µg
Stainless Steel	Visual	-	+	+	+	+	+
	ELISA	-	-	-	+	+	+
	Sensitive ATP	-	-	+	+	+	+
	Total Protein	-	+	+	+	+	+
Urethane-faced	Visual	-	-	-	+	+	+
	ELISA	-	-	-	+	+	+
	Sensitive ATP	-	-	+	+	+	+
	Total Protein	-	+/-	+	+	+	+
Teflon	Visual	-	-	+	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	+	+	+	+	+
	Total Protein	-	+/-	+	+	+	+

Results

Table 2. Comparison of Detection Methods for Egg Residue on Food-Contact Surfaces

Surface	Method of Detection	Amount of Spray-Dried Egg Spotted on Surface					
		0 µg (control)	10 µg	50 µg	100 µg	500 µg	1000 µg
Stainless Steel	Visual	-	+	+	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	-	-	+	+
	Total Protein	-	+	+	+	+	+
Urethane-faced	Visual	-	-	-	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	-	-	+	+
	Total Protein	-	+	+	+	+	+
Teflon	Visual	-	-	-	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	-	-	+	+
	Total Protein	-	+/-	+	+	+	+

Results

Table 3. Comparison of Detection Methods for Peanut residue on Food-Contact Surfaces

Surface	Method of Detection	Amount of Peanut Flour Spotted on Surface					
		0 µg (control)	10 µg	50 µg	100 µg	500 µg	1000 µg
Stainless Steel	Visual	-	-	-	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	-	+	+	+
	Total Protein	-	-	+	+	+	+
Urethane-faced	Visual	-	-	-	+	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	+	+	+	+
	Total Protein	-	-	+	+	+	+
Teflon	Visual	-	-	-	-	+	+
	ELISA	-	-	-	-	+	+
	Sensitive ATP	-	-	-	+	+	+
	Total Protein	-	-	+	+	+	+

ATP positive >181000 RLU on SS plates
ATP positive >180,638 RLU on Teflon plates
ATP positive>145,704 RLU on Urethane plates
ELISA positive ≥ 5ppm
Total protein positive ≥ 10-20 µg

- In general, the detection limits (DL) for visual inspection were consistent with those determined by ELISA for all foods tested.
- ELISA and visual tests detected milk at 100 µg on all food surfaces, but 10-50 µg when tested by sensitive ATP and total protein swabs (Table 1).
- The DL (visual, ELISA and ATP) for egg was 500 µg on all food contact surfaces, but lower with total protein swabs (Table 2).
- The ELISA and visual detection limit for peanut was 500 µg on all food surfaces, but 50-100 µg when tested with the sensitive ATP and total protein swabs (Table 3).
- In some cases, total protein swabs detected food residues at or below levels detected by sensitive ATP swabs.

Significance

This study shows that visual inspection and analytical methods are valuable tools for detecting the presence of allergenic foods on food-contact surfaces, but limitations exist and care must be taken when choosing a method for detecting the presence of food residues on food-contact surfaces.

References

Taylor, S.L., Hefle, S.L., Farnum, A.K., Rizk, S.W., Yeung, J., Barnett, M.E., Busta, F., Shank, F.R., Newsome, R., Davis, S., and Bryant, C. 2006. Analysis and evaluation of food manufacturing practices used to address allergen concerns. *Comprehensive Reviews in Food Science and Food Safety*, Volume 5, pp. 138-157.

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