

**Optimizing sample plans to improve microbiological safety  
in a food processing plant**

Hassan M. Masri

Thesis submitted to the Faculty of the Virginia Polytechnic Institute and State University  
in partial fulfillment of the requirements for the degree of

Master of Science in Life Sciences  
In  
Food Science and Technology

Joseph D. Eifert, Committee Chair  
Renee R. Boyer  
Hengjian Wang

June 7, 2013  
Blacksburg, Virginia

Keywords: sampling plan, pathogen, low-moisture food, environment

# Optimizing sample plans to improve microbiological safety in a food processing plant

Hassan M. Masri

## ABSTRACT

*Salmonella* and *Cronobacter sakazakii* are two leading causes of foodborne illness associated with low-moisture foods, including infant formula. Both causative organisms can persist in food manufacturing processing environments and contaminate finished product if programs are not in place to limit their introduction and control their spread. An environmental sampling and monitoring program is an important tool that food manufacturers use to determine the effectiveness of their sanitation practices and pathogen control efforts. Guidance for initiating an environmental sampling plan and evaluating the plan is lacking.

The objective of this study was to develop microbiological environmental sampling plans based on the answers to a series of questions related to product hazards, processing risks and controls, and knowledge of appropriate microbiological sampling and testing protocols. Furthermore, these initial sampling plans were related to the volume of product and size of the processing facility. An interactive spreadsheet tool for designing sampling monitoring plans for an infant formula process was developed using Microsoft Excel.

Additionally, the tool can be used to record qualitative and quantitative sample test results, and to alert the user how the upcoming sampling plan will be changed, if necessary, based on monthly test summaries. The sampling tool provides a simple method for selecting an appropriate environmental sampling plan (samples per zone per month) and provides a rationale and guidance for creating and modifying these plans. Effective sampling plans and trend analysis of sample test results support the food processors decisions for implementing controls to enhance food safety.

## **DEDICATION**

I dedicate my thesis work to my family and many friends. A special feeling of gratitude to my loving parents, Mohammed Masri and Ameerah Al Mashat, whose words of encouragement and push for tenacity ring in my ears. I also dedicate this thesis to my many friends who have supported me throughout the process. I will always appreciate all they have done, especially Mohammed Al Shuniaber for helping me study and pass my classes. I dedicate this work and give special thanks to my wife Hawazen Nadrah and my wonderful daughter Ameerah Masri for being there for me throughout the entire masters' program. Both of you have been my best cheerleaders.

## **ACKNOWLEDGEMENTS**

I would like to express my gratitude to my supervisor Dr. Joseph Eifert for the useful comments, remarks and engagement through the learning process of this master thesis.

I would like to thank my family- my mother Ameerah Al- Mashat, my wife Hawazen Nadrah and beloved daughter Ameerah Masri who supported me during my years of study. As well as my friends Mohammed Al Shuniaber, Matthew Schroeder, Ivan Volonsevich and others for the motivation that inspired me. I would like to thank the Saudi Arabia Cultural Mission for the scholarship and financial support granted to me during my graduate life. And, thanks to Virginia Tech and the Department of Food Science and Technology for the opportunity to be one of their team and family.

## TABLE OF CONTENTS

INTRODUCTION	1
LITERATURE REVIEW	4
A. Microbial Sampling and Testing in Food Manufacturing	4
B. Microbial Sampling in Milk Powder Processing to Enhance Food Safety	5
1. Milk powder production	9
2. Infant cereal and formula production	6
3. Foodborne illness associated with dried milk products:	7
a. <i>Salmonella</i>	8
b. <i>Cronobacter sakazakii</i>	9
4. Control of <i>Salmonella</i> and <i>C. sakazakii</i> in low-moisture foods	10
C. Microbiological Environmental Sample Selection	11
1. Zoning in a processing environment:	11
2. Sample type and frequency	12
3. Number of sample sites	13
4. Sample analyses	14
D. Sampling Plans in Food Manufacturing	14
1. Sampling plan rationale	16
2. Sampling plans for monitoring product and process safety and quality	18
a. Attributes sampling plans:	19
b. Variables sampling plans	19
3. Sampling for routine inspection	20
4. Investigational sampling	22
5. Tightened inspection/ skip lot sampling	23
E. Development and Evaluation of Environmental Sampling Plans	23

MATERIALS AND METHODS	25
A. Development of an Interactive Tool to Establish an Environmental Sampling Plan for an Infant Formula/ Infant Cereal Processor	26
1. Initial sampling plan based on product and process food safety risk	26
2. Questionnaire to determine relative product/process risk	27
a. Step One: Food safety procedures	27
b. Step Two: Processing hazards	28
c. Step Three: Microbiological Sampling and Testing	28
3. Questionnaire to determine relative production volume/ plant size	29
4. Question response scoring	30
5. Sampling plans based on process risk and production volume	31
B. Development of a Spreadsheet Tool to Record and Summarize Environmental Sample Test Results for an Infant Formula/ Infant Cereal Processor	32
1. Recording environmental sample test results	32
2. Sample Test Result Summaries	33
C. Sampling Plan Modification based on Monthly Evaluations of Test Result	34
RESULTS	37
DISCUSSION	40
SUMMARY	44
REFERENCES	59
APPENDICES	
A. Sampling Test Improvement Workbook	63

## LIST OF FIGURES

Figure 1. Questions related to product and product risk determination (food safety procedures, processing, sampling /testing).	45
Figure 2. Questions related to relative production volume.	46
Figure 3. Current sampling plan view describes the sample plan category code, risk level, current guideline state, production volume level, data entry and data report view.	47
Figure 4. Sample site view describes site code, zone, sample description or location.	48
Figure 5. Summary report pivot table for both enterobacteriaceae “EB” count and <i>Salmonella</i> “S” test result, including zone, total number of samples and site location	49
Figure 6. Sample data entry window includes (date sample taken, date sample analyzed, sample zone, sample site code, and enterobacteriaceae “EB” count and <i>Salmonella</i> “S” test result.	50
Figure 7. Initial sample plan software interface.	51

## LIST OF TABLES

Table 1. Summary of question response scores required for each of the 9 possible starting environmental sampling plans	52
Table 2. Summary of total environmental samples required, and maximum number of unacceptable (failed) samples, for each of the 9 possible starting environmental sampling plans.	53
Table 3. Data collection report view includes date sample taken, date sample analyzed, sampling zone, sample site code, enterobacteriaceae “EB” test result, <i>Salmonella</i> “S” test result, and sample test result validation (Pass/Fail).	54
Table 4. Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 1 with guideline for adjusting sample plan totals by zone (“if # of failed <i>Salmonella</i> samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, until you receive a negative result for 2 weeks”).	55
Table 5. Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 2 with guideline for adjusting sample plan totals by zone (“If # failed samples in a month > than limit, then increase EB samples by 2X each zone for 1 month until you receive under norm results”).	56
Table 6. Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 3 with guideline for adjusting sample plan totals by zone (“if # of failed samples > limit for 3 consecutive months, then shift to higher risk sampling plan (low --> med., or med. --> high) for 1 months till you receive under norm results”).	57
Table 7. Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 4 with guideline for adjusting sample plan totals by zone (“if # of failed samples < limit for 3 consecutive months, then you may reduce total # of sample analyses by 10% (by pooling samples) if you have a good history of your results”).	58



## INTRODUCTION

The annual number of foodborne illnesses in the United States is estimated at 49 million cases, and *Salmonella* bacteria are responsible for more than 1 million cases of illness each year (Sallan et al., 2011). The medical care and lost productivity cost for *Salmonella* between \$6.5 and \$34.9 billion annually (IFT, 2004). *Salmonella* is a generic name applied to a group of approximately 2,000 biochemically related serotypes responsible for foodborne illness. *Salmonella* can cause illness with an infectious dose as few as 15 cells (IFT, 2004).

A significant food safety risk may occur in contaminated foods produced as ready-to-eat with no additional *Salmonella* kill step in the process. This type of product includes low moisture products that do not support *Salmonella* growth. However, low cell counts of *Salmonella* in foods can cause illness, and the presence of this organism in low moisture ready-to-eat foods must be prevented. A number of outbreaks of salmonellosis have been associated with the consumption of ready-to-eat and low-moisture products, including chocolate, powdered infant formula and, more recently, peanut butter. Although foodborne illness outbreaks are rare due to *Salmonella* from low-moisture products, they often impact large numbers of people (GMA, 2009).

Another pathogen associated with low-moisture foods is *Cronobacter sakazakii*. *Cronobacter* spp. infection has been associated with powdered infant milk formula, and several voluntary recalls of powdered formula have been issued because of contamination by this pathogen (Norberg et al., 2011).

Control of these pathogens in low-moisture foods often requires sampling and testing of the product, process environment or both. Many microbiological tests are available to detect these pathogens directly or other indicator organisms such as

Enterobacteriaceae. Most pathogen tests for *S. enterica* and *C. sakazakii* are qualitative since we are usually expecting an absence of these organisms in foods and the process environment. Tests for indicator organisms or microbial tests to verify sanitation are often quantitative.

The food industry has many specific tools and procedures for conducting microbiological tests on food samples or the food process environment. Unfortunately, procedures or guidance for selecting appropriate environmental samples is lacking. Very little guidance is available that specifically describes appropriate sample sizes, numbers, frequency, type, location, etc. Food industries often design microbiological sampling plans that can range from excessive to minimal since they may not have a rationale for creating a sampling other than cost, convenience or caution.

Technology provides us with suitable analytical tools which can assist us in developing a new environmental monitoring sampling plan module that can be applicable to several food commodity processes. Designing a monitoring sampling plan software program with consideration of multiple factors of the operation will ensure the adequacy of the collected environment samples. One aim of this study is to gather and analyze the collected data from the process line over a period of time, on a statistical basis.

Another step is to determine accept and reject criteria of the tested environment samples. The limit of those criteria must be established and evaluated periodically. A written environmental sampling monitoring program is imperative. The significance of the environment samples collected from the production line depends on the zone classification, type of cleaning and the exposure of the product. Designing an

environmental sampling monitoring program tool based on risks associated with processing in a specific facility and the ability to control those risks can help food processors carry out this important food safety program. The tool could assess the food manufacturers' risk that can pose threat to the food during the production cycle time.

Adapting this type of tool can support the decision of releasing the finish product or control the activity in the high risk operation area. Furthermore, sampling plans should be modified at regular intervals, based on analysis of previously collected samples, to facilitate the detection and control of target microorganisms.

Data collection is significant to the processor to determine the weak point and what major controls must be consider improving the design tool program. Trends of collected samples can be analyzed and decisions can be made to restrict the control level of the production line from high to low. Based on the data collected from the sample test results, the quantity and frequency of future samples can be changed. In other words, processors could have a way to justify an increase or decrease in the number of environmental samples collected based on previous test results.

## LITERATURE REVIEW

### A. Microbial Sampling and Testing in Food Manufacturing

The food processing industries can establish checks on their food products, food processes or both to ensure the safety and quality of the food they produce. For example, lot acceptance sampling and testing programs and environmental sampling and testing programs can be used to enhance food safety, but these programs differ in many respects. An environmental sampling program can be used to monitor and verify whether a processor's Good Hygienic Practices (GHP) are effective and being correctly applied to prevent unacceptable contamination. GHP's can be an effective way to control certain pathogens and spoilage microorganisms within the process environment and to prevent contamination of foods and then estimate whether food is acceptable.

A plan to sample the plant environment can be based on the information needed by the processor, the layout of their process and processing plant, and by their previous experience with sampling and testing the environment. A variety of sample locations or sites may be relatively straightforward to select. The number and frequency of samples; how, when and where they are collected; how they are handled between sampling and analysis and the sensitivity of the analytical method are all crucial to accomplish the goals of the environmental sampling program. The cost of the environmental sampling program is one limiting factor that manufacturers must address (Tompkin, 2004).

Microbiological samples of finished product, in-process product or the plant environment can be analyzed qualitatively and/or quantitatively for pathogenic

microorganisms, spoilage microorganisms, microbial indicators of microorganisms of concern or microbial indicators of insufficient sanitation.

The objective of the sampling plan should be defined before choosing the sampling plan. Ensuring the complete absence of the defect cannot be defined by a sampling plan. When a food safety objective or other limit exists, choosing a sampling plan will be easy. The stringency of the sampling plan then can be determined to detect the defect levels. The potential source of the problem and the population to be sampled must be determined when selecting a sampling plan. Developing an appropriate tool to collect samples can be meaningful to gain information (ICMSF, 2002).

When data is acquired and frequently reviewed, the environment sampling plan can be useful. Reviewing data from the recent and past test results taken from the processing environment to detect weaknesses and trends which might be evident are helpful. Data from environmental sampling tests result within the acceptance limits can support the decision that a normal routine level of sampling be continued. The reason should be determined if an increased risk of contamination is indicated on trend or other information. The detection of the contamination and its causes is the goal of the intensified sampling program (ICMSF, 2002).

## **B. Microbial Sampling in Milk Powder Processing to Enhance Food Safety**

### **1. Milk powder production**

Raw milk obtained from cows undergoes a wide variety of technologies and processing to be included in commodities such as fluid milk, cheese, fermented milks and milk powder. Using appropriate technologies on raw milk such as spray drying or

roller drying can produce many products of milk including whole milk, skimmed milk and cream (ICMSF, 2011).

These dried milk products can be used either directly after reconstitution or as an ingredient in many products. Infant formula and infant cereals are produced using the same technologies and process line usually but differ in regulatory requirements (ICMSF, 2011).

## **2. Infant cereal and formula production**

Infant cereals are made from one or more grains, usually rice, oat or wheat, and supplemented with calcium, iron, and vitamins. These cereals are sold as a dry or flaked product that must be reconstituted with water or milk. Many of these cereals are developed for babies between 6 to 12 months in age. Examples of such foods include cereal based infant foods, uncooked breakfast cereals, or products designed to be baked at home (ERS, 2010).

Infant formula can be manufactured in many forms like ready-to-feed ultra-high temperature products or concentrated sterilized products. Infant formula can be manufactured in three process types (ICMSF, 2011):

- 1- Wet mix process: which all the unprocessed ingredients are separately handled in a liquid form, which then be heat treated and dried before filling stage is completed.
- 2- Dry mix process: which all separately processed ingredients are dry blended before the filling stage.

3- Combined process: which under the wet mix process both unprocessed raw material and part of the ingredient are processed to form a base powder.

In manufacturing infant cereal a cereal soup is heated before further processing. After the heating step is completed the cereal soup is transferred to the roller-dryer. In this step the cereal soup is evenly distributed in a thin film on the roller-dryer drum. Powder or small flakes are then obtained from the cereal film to form a base powder. The obtained base powder from the previous step would be used directly or mixed with other dry ingredient such as vitamin, fruit or vegetable flaks and powder (ICMSF, 2011).

### **3. Foodborne illness associated with dried milk products:**

Outbreaks have been linked to dried milk products including infant formula and cereals in many countries. In some cases addition of hydrating ingredients such as water or milk can assist the growth of bacterial pathogens. Powdered infant formula is not a sterile product and can get contaminated with pathogens. Correct preparing and handling of the product can reduce the risk of illness. Manufacturing commercially sterile powder infant formula is not feasible with the current processing technology. During the production of powdered infant formula it can become contaminated with harmful bacteria, such as *Salmonella enterica* and *Cronobacter sakazakii* (WHO, 2007). Currently these are the primary bacterial pathogens of concerns in this product.

a. *Salmonella*

*Salmonella* spp. are Gram-negative, rod-shaped motile bacteria (non-motile exceptions *S. gallinarum* and *S. pullorum*), and is non-spore forming (FDA, 2012). *Salmonella* is a generic name applied to a group of approximately 2,000 biochemically related serotypes responsible for millions of cases of foodborne illness worldwide. The number of foodborne cases caused by salmonellosis annually is roughly 0.6 to 1.7 million in the United States (Scallan et al., 2011). Salmonellosis is a self-limiting gastroenteritis which may be misdiagnosed as intestinal influenza by the patient or the physician, and therefore it is grossly underreported (IFT, 2004). *Salmonella* are recognized by two clinical indicators, enteric fever and foodborne illness syndrome (IFT, 2004). The enteric fever (a severe, life-threatening illness) is commonly referred to as typhoid fever, it is primarily caused by *Salmonella* Typhi. Foodborne illness syndrome is usually caused by *Salmonella enterica*.

The microorganisms responsible, in both cases, enter the body via the oral route. Typically, *Salmonella* infection resulting from foodborne illness is commonly characterized by a self-limiting acute gastroenteritis. The usual but not the only common vehicle of *Salmonella* contamination is food or water. *Salmonella* infectious dose is as few as 15 (IFT, 2004).

*Salmonella* can enter the food supply in multiple ways (IFT, 2004): 1) food animals can harbor *Salmonella*, making meats, poultry, eggs, and milk often implicated vehicles for *Salmonella*; 2) *Salmonella*, which is introduced into the environment possibly through manure and litter, may survive and contaminate fruits and vegetables on the farm; and 3) cross-contamination in the food service environment or the home,



often between raw poultry and ready-to-eat products, such as raw vegetables, can also cause salmonellosis.

Cross contamination is an important concern with this pathogen, since it is one of the ways for *Salmonella* to enter the food supply especially with food that does not require a further kill step before consumption, such as some low-moisture product. A significant food safety risk may occur when this transfer takes place where the product is ready-to-eat with no additional *Salmonella* kill step in the process. This type of product includes low moisture products that do not support *Salmonella* growth. However, low cells counts of *Salmonella* in foods can cause illness, and the presence of the organism in low moisture ready-to-eat foods must be prevented. A number of outbreaks of salmonellosis have been associated with the consumption of ready-to-eat and low-moisture products, including chocolate, powdered infant formula and more recently peanut butter. Although *Salmonella* outbreaks are rare from low-moisture products, they can impact large numbers of people (GMA, 2009).

b. *Cronobacter sakazakii*

*Cronobacter sakazakii*, formerly *Enterobacter sakazakii*, is a Gram-negative, motile, rod-shaped, non-sporulating pathogenic bacterium that can cause foodborne illness, primarily among infants and immunocompromised adults. The organism is able to survive in low-moisture foods, such as powdered infant formula, for long periods (FDA, 2012). For children infected with *C. sakazakii*, 50% are less than 1 week old and 75% are less than 1 month old.

*Cronobacter* infections were reported in immuno-compromised and elderly adults (FDA, 2012). Typically, contaminated powdered infant formula products with *Cronobacter* are the common source for infections (FDA, 2012). Powdered infant formula manufacturers worldwide are threatened by *Cronobacter* spp; therefore, it is important to understand the factors that support the survival and the growth of this pathogen. *Cronobacter* spp are tolerant to high temperature and low water activity which make it difficult to be controlled. *Cronobacter* spp. stress tolerance appears to be dependent on the strain and growth phases (Norberg et al., 2011). *Cronobacter* contamination can occur after the heat treatment pasteurization step used for the powdered milk (FDA, 2012). This indicates poor hygiene practices after the pasteurization step is the source of *Cronobacter* spp. contamination (Norberg et al., 2011).

#### **4. Control of *Salmonella* and *C. sakazakii* in low-moisture foods**

In low moisture foods, the barrier to grow vegetative pathogens, including *Salmonella* spp is water activity ( $A_w$ ) (GMA, 2009). Water activity ( $A_w$ ), is defined by the ratio of water vapor pressure of food to the vapor water pressure of pure water at a specific atmosphere. Moreover, low moisture products are characterized as a low water activity product which does not support the growth of *Salmonella*. Products which are characterized as low moisture products are powdered milk products, chocolate, peanut butter, infant formula and toasted cereal. The presence of *Salmonella* in low-moisture products is a concern because low numbers of *Salmonella* in foods can survive and cause illness. This is contrary to a common misconception that low numbers of

*Salmonella* are not a problem in low-moisture foods because these products do not support *Salmonella* growth. Although these products do not support the growth of *Salmonella*, all have been implicated in outbreaks of salmonellosis. After investigating the outbreaks it was suggested that cross contamination plays a major role in the contamination by *Salmonella* of these products (GMA, 2009).

### **C. Microbiological Environmental Sample Selection**

An environmental sampling plan can be designed to optimize the detection and control of undesirable microorganisms. The plan should include specific sample information such as type, quantity, frequency, location, analysis, etc. Additionally, strategies to design these sampling plans for food processing environments should consider the following (Tompkin, 2004):

- Control the condition which can lead to creating biofilms. This approach may require redesign of equipment to use appropriate material.
- Establish an effective environmental sampling program. The need of this type of program is to detect any pathogenic contamination in a timely manner.
- Short term assessment of the results.
- Longer term review of the data quarterly or annually.

#### **1. Zoning in a processing environment:**

An environmental sampling program must include various areas throughout the production process. Dividing the production process line into zones is the simplest way to design an effective environmental sampling program. Based on the production facility

and process line specification, sampling sites from each zone can be determined. To improve the control of *Salmonella* a zoning concept must be established (Marriott and Gravani, 2006). The zone concept is based on four sampling areas and sample types which can aid the identification of contamination spots:

Zone 1— includes the direct contact surfaces. These areas include equipment utensils, and containers with direct contact with food products.

Zone 2— area with indirect contact with food in the facility such as equipment parts or other surfaces that personnel may come in contact with near Zone 1. This includes drains, utility pipes, and heating, ventilation or air conditioning system equipment.

Zone 3— area with less contact to food in the facility unlike Zone 2. That includes floors, walls, and other items in contact with floors, walls, cleaning equipment.

Zone 4— includes maintenance equipment and areas further away from production such as hallways and entrances of the facilities. Some food processors may group Zone 3 and Zone 4 samples together when designing a three zone sampling plan.

## **2. Sample type and frequency**

Each processing facility must select appropriate sample types and a frequency of sample collection that provides them sufficient information to maintain or improve the level of plant hygiene or reduce the presence of pathogenic bacteria. Samples that are typically collected for microbiological analysis include raw products or ingredients, equipment surfaces, processing water, walls, floors, drains and air. For each sample,

the time of collection, location sampled, sample type, sample quantity, and analysis required must be specified.

The frequency of sample collection for specific sample types or locations can be based on several factors including traffic patterns in the plant, production volume, sanitation procedures and frequencies, previous history of sample analysis data, and microbiological guidelines or action levels. The frequency of sample collection can vary for different plant locations or surfaces.

### **3. Number of sample sites**

For site qualification, the International Organization for Standardization (ISO) standard 14644-1 describes a method to determine the number of sampling sites (Sutton, 2010). The minimum number of sample sites should be determined by the following equation (Sutton, 2010):

$$N_L = \sqrt{A}$$

Where:

$N_L$  is the minimum number of sampling locations (rounded up to a whole number)

$A$  is the area of the clean room or zone in meters<sup>2</sup>.

Alternatively, food processors may choose other methods to determine an appropriate number of samples to collect for their sampling plan. Their choices may be based on the number of processing lines in their plant, the number of hours or days per week that they process products, regulatory guidelines, previous sampling history, and, of course, cost and convenience.

Protocols for the environmental sampling plan may not be designed on a statistical basis. Thus, sampling plans are based on experience or knowledge of the site to detect any failure on GHP. When evidence indicates increased risk of contamination the number, timing, frequency and sampling sites may be increased (ICMSF, 2002).

#### **4. Sample analyses**

Sampling and analytical tests may be conducted for specific pathogens such as *Salmonella* or *Listeria monocytogenes*. Alternatively they can target indicators of pathogen presence such as tests for *Listeria* spp. to demonstrate the possibility that *Listeria monocytogenes* is present or tests for Enterobacteriaceae that may indicate the presence of *Salmonella*. Also, aerobic plate counts and ATP bioluminescence assays are often used to determine areas that need additional cleaning and sanitation.

#### **D. Sampling Plans in Food Manufacturing**

An environmental sampling and monitoring program can include numerous locations throughout the process line. The collected samples can also vary from spilled product on the floor or equipment, vacuum cleaners, floors, walls, and other surfaces. And, the type of collected sample may vary, as well, from solid to liquid samples. Locations where samples must be collected from differ depend on their proximity to the food products and process. The area with high exposure of the product will be sampled more. The frequency of the samples collection may vary to ensure that the hygienic requirement is met during the production cycle. Conducting this type of monitoring program should include: sample type, sample location, sampling time and frequency,

sample quantity, and procedures for sample collection and analyses. After establishing an environmental sampling monitoring program, it must be verified and validated from the processer owner. The monitoring program must be evaluated periodically to ensure the adequacy of it met the goal of sustain hygienic production environment.

Food manufacturers are required to follow guidelines and adopt control measures to ensure that the food produced from their facility is safe. Compliance with food safety regulations and adopting appropriate regulatory guidelines is the first step in producing a safe food. Procedures and policies implemented on site in each food manufacturing establishment differ from another. During production it is crucial to comply with the control measure in place to ensure the safety of the food. Evaluating those measures periodically and the risk to the finish product is important as well.

Producing food in a sanitary environment is an important factor as the risk of cross contaminating the food will reduce. Implementing a food sampling monitoring program is essential. Under those monitoring program any part of the process can be included. Example: incoming material, online samples, equipment's, tools, floor, walls and the finish product.

As a normal practice, most of the food manufacturers, following the governmental regulations or guidance, design their own monitoring sampling plan which is applicable to their specific production line. Therefore, individual environmental monitoring sampling plan is established by each processer with consideration of compliance with government regulation.

Food manufacturers are required to comply with a regulatory authority in the means of releasing a safe finish product to consumer. The type of evidence may vary

from process control documents, online inspection, a product or environmental sampling plan, or microbial test results. Moreover, food manufacturers are required to prove the adequacy of the production line. Adapting several control methods such as hazard analysis critical control point (HACCP) programs, environmental zoning concept, environment sampling monitoring program and good manufacturing practices (GMP) would support the decision of whether they are capable of releasing a safe product.

These types of control measures in place influence the readiness of the food manufacturing facility to produce safe food. Conducting a study on the posed hazards from the production line within the food manufacturing facility is necessary. These study questions must answer: What food am I producing? Where am I producing the food? What are the hazards of concern to my food? And many other factors must be considered, all which reflect on the control implemented to ensure the safety of food produced by the manufacturer.

### **1. Sampling plan rationale**

A sampling plan is an executable plan of action that addresses the sampling and analytical requirements of a specific situation and adheres to the specific sampling strategy. The sampling plan must specify the sampling approaches, methods, and analyses, as well as the number, types, and locations of samples to be collected in a given physical space. The sampling plan should account for the area under consideration, the number of samples, and the collection locations needed for statistical confidence as determined by directed and/or statistical sampling designs.



Directed sample collection utilizes an expert in the field to determine the suitability of the plan to meet the experimental objectives. Statistical sampling utilizes a mathematical framework to determine if the number and location of sample collection sites meets specific characterization objectives (NIST, 2012).

A sampling plan can vary widely in its goal. The objectives of a sampling design for data collected from the environment are (EPA, 2002):

- Support a decision whether the contamination level exceeds a threshold unacceptable risk.
- Determine whether the characteristics of two populations differ by some amount.
- Estimate the mean characteristic of population with the same interest.
- Identify locations having high level of contaminations.
- Characterize the extent of contamination at site.
- Monitor trend of the environment condition.

To ensure that resulting data are adequately representative of the target population, a well-planned sampling design is made. Efficient use of time, money, and human resources are critical considerations for sampling design process. Minimum costs of resources should meet the needs of the study of a good sampling design (EPA, 2002). A number of samples and identifications of the particular samples are indicators of a complete sampling design. Moreover, a complete sampling design will include an explanation and justification for the number and the positions/timings of the samples (EPA, 2002):

Detecting pathogens in foods was thought to be strictly outside the scope of environmental sampling. A food manufacturing facility can potentially become cross contaminated from environmental pathogens so we will consider briefly this aspect of pathogen sampling. A food processing facility is assumed to follow the principles of good hygienic practice (GHP) as outlined by Codex (1997). Leading us to presume that there are procedures in place to manage the risk of environmental pathogens by: 1) minimizing cross contamination from raw materials to finished product; 2) having equipment of suitable sanitary design; 3) appropriately maintaining and sanitizing equipment; 4) removing waste; and 5) training personnel. Then, to ensure that those procedures are working an environmental sampling program must be implemented (Legan & Vandeven, 2003).

With regards to their statistical background and in relation to other risk management approaches such as HACCP or Food Safety Objectives, microbiological criteria and sampling plans are not fully understood. Microbiological test performed on several sample units is a simple way to decide whether to accept or reject a food lot (Dahms, 2003).

## **2. Sampling plans for monitoring product and process safety and quality**

To ensure food quality and safety, two sampling plans are used: attributes sampling plans and variables sampling plans. These types of microbiological testing are used to make decisions concerning the safety or quality of foods (Dahms, 2003).

Attributes plans are used to evaluate qualitative data (presence or absence of analyte)

or quantitative data that have been grouped (e.g., <10 cfu/g, 10 to 100 cfu/g, >100 cfu/g). Non-grouped quantitative data can be evaluated with variables plans.

a. Attributes sampling plans: Attribute sampling, also known as proportional sampling, allows one to measure the probability of discrete possible outcomes. The presence or absence of a specific contaminant is an example of a discrete outcome that could be measured with attribute sampling. There are two methods of attribute sampling plans qualitative two-class attribution and quantitative three-class attribution test (Dahms, 2003). For the qualitative test of presence or absence of the pathogens, a two-class plan is defined by two numbers for decision making process. First, denote  $n$ , which determines the samples units independently and randomly picked from a production lot. Secondly, denote  $c$ , which is the maximum acceptable number of samples yielding unsatisfactory result. In the case of quantitative grouped data applied to a two-class plan, there is one microbiological limit denoted  $m$ , which separates acceptable from defective quality.

Moreover, an Operating Characteristic (OC) curve is used to visualize the performance of the sampling plan. The OC curve has two scales: a horizontal scale showing the percentage of positive units in the lot being tested, and a vertical scale giving the probability of acceptance.

b. Variables sampling plans: Variable sampling allows one to measure quantities. For examples, variable sampling could be used to measure how many bacteria are present at a given time, or temperatures of a processing area at defined time intervals. When decisions are not based on qualitative analytical tests, quantitative analytical tests

are applied, working with data grouped according to a single microbiological logical limit  $m$ . Three-class plans are used where the quality of food is divided into three attribute classes. In two-class plans samples results based on quantitative analytical results above a concentration  $m$  that in a three-class plan separate good quality from slightly acceptable quality but a certain number donate as  $c$ , can be accepted. However, sample test results exceeding the second microbiological limit  $M$  are rejected if any test result of  $n$  sample unit is above  $M$ . Therefore, three-class plans OC curves result in three dimensional graphs which are difficult to compare with two dimensional OC curves.

### **3. Sampling for routine inspection:**

To ensure that the operation remains under control by detecting any increase in the risk of cross-contamination, routine environmental samples must be collected. A normal risk of cross-contamination, when the operation is under control, first must be determined by:

- Selection of one pathogen or indicator organism for the ongoing monitoring.
- Take samples at various stages of the process flow, to determine the microbiological condition facility.
- Residues of food samples can be collect as monitoring samples.

Environmental sampling protocols are designed to pay most attention to those areas known to pose the highest risk of product contamination and they are not statistically-based. However, some statistical concepts are applicable. Assigning number of zones within the processing plant is common practice, where different zones have different

levels of risk of contaminating the product. Zoning concept is defined in four zones as mentioned previously. Based on experience of the sites the exact balance of samples from the four zones is most likely to indicate that the operation is 'out of control' in terms of good hygienic practice (Legan & Vandeven, 2003).

The sampling protocol and improved knowledge of the operation will over time shift the selection sites. Knowing where to sample as well as when to sample are important. The most critical time may be immediately after startup in some operations. Special sampling can be implemented in response to known risk factors such as construction the frequency of sampling may be increased if evidence indicates an increased risk (Legan & Vandeven, 2003).

The Grocery Manufacturers Association (GMA, 2009) included an example of an environmental monitoring program for production of low-moisture foods in their guidance document on *Salmonella* control in these products. The GMA document details a monitoring program with 4 sampling zones where samples are tested for *Salmonella* primarily in zones 2, 3, and 4. The number of samples in each zone decreases as you move from zone 2 to zone 3 and zone 4. Tests for indicator organisms such as Aerobic Plate Count or Enterobacteriaceae are recommended for zone 1 (product contact surfaces in primary control areas). They note that Enterobacteriaceae is a useful indicator of process hygiene and may be monitored in parallel as a hygiene indicator for verification of general sanitation effectiveness. However, it cannot be a substitute for the direct monitoring of *Salmonella* because, while high levels of Enterobacteriaceae suggest an increased risk for the presence of

*Salmonella*, low levels of Enterobacteriaceae do not guarantee the absence of the pathogen (EFSA, 2007; Cordier, 2008).

Another example of sampling plan guidance was recently published by the US Dept. of Agriculture, Food Safety and Inspection Service for use by their Enforcement, Investigations and Analysis Officers (EIAOs) to follow when collecting product samples during Intensified verification testing protocols for sampling of product, food contact surfaces, and environmental surfaces for *Listeria monocytogenes (Im)* or *Salmonella*. Effective in 2013, samples collected for the *L. monocytogenes* program will consist of sampling units of 10 food contact surface, 5 environmental (non-food contact surface), and 5 ready-to-eat (RTE) product samples. Additionally, when sampling for *Salmonella*, EIAOs are to collect 5 food contact surface samples, 8 environmental samples, and 5 RTE product samples. The instructions provide some guidance on when to collect the samples during a day, and suggest locations to sample, but they do not state the quantity and frequency of sample collection at each sample site (USDA, 2013).

#### **4. Investigational sampling:**

Information suggesting that a problem already exists can lead to an investigation response. To correct the problem the source must be known first. To investigate a problem efficiently, a random sample must be obtained and knowledge must be applied to: 1) microbiology; 2) process operations, 3) equipment design; 4) information gained from visual inspection of the operation; and 5) sampling sites most likely to harbor the organism(s) of concern. It is important not to jump to conclusions, even at the same time as applying pre-existing knowledge, mainly when resources are limited and time is

constrained. Investigational sampling is very likely to be repeated to maintain a degree of flexibility. In more detail the influences between the last 'point of absence' and the first 'point of detection' can be examine. The investigation work over time can sometimes be very lengthy towards identifying the source of contamination (Legan & Vandeven, 2003).

### **5. Tightened inspection/ skip lot sampling**

With an ongoing relationship with a supplier, or other ongoing sampling situation, a level of confidence in the performance of that supplier over time can be developed. Skipping sampling of some lots altogether and using the freed sampling resources where they can be more beneficial that may lead us to relax the rate of sampling. However, if a defective were detected the initial sampling rate will be reverted or even more stringent, until the confidence of the supplier is developed again to an acceptable level (Legan & Vandeven, 2003).

## **E. Development and Evaluation of Environmental Sampling Plans**

The response to information that indicates a problem is a major limitation throughout the food industry. This can be due to failure to organize the results in a manner that facilitates review (Tompkin, 2004). For example:

1. Failure to recognize an evolving problem or its significance,
2. Simply filing results without review or,
3. Finally, an individual or group is not assigned responsibility or held accountable for identifying and responding to a problem.

Time needed and the difficulty to detect the source of contamination is a limitation in industry's response. All samples should be analyzed individually rather than pooled, and, samples should be collected more frequently and additional sites should be included when striving to detect the source. Dates and times when positive results have occurred should be easily traced to a location on the map showing the layout of equipment's in the room and sites. This can be demonstrated in the following order (Tompkin, 2004):

1. Do the results reveal patterns with certain equipment showing more positives?
2. Where in the flow of food through the process do the first positives occur?

In general, the microorganisms flow downstream from the source of contamination with the food. Identifying the source and the pathways of contamination can be determined by fingerprinting isolates (Tompkin, 2004).

Frequently, food processors react to unacceptable test results through additional sampling or sanitation procedures. However, continual, long-term evaluation of environmental sampling plans and test results should be performed to determine if there are trends in microbial detection. The evaluation of the sampling plan and the test data over extended times may lead to changes in the number of samples collected, test sample frequency, location and analysis performed, or in the plant's corrective actions. A thorough evaluation of the data can lead to increased sampling for potential problem areas and decreased sampling frequencies for areas that have generally negative test results.



## MATERIALS AND METHODS

An interactive spreadsheet tool for designing sampling monitoring plans, that suggest one of 9 basic sampling plans based on product/ process risk (3 levels) and production volume (3 levels), was developed using Microsoft Excel 2010 (Microsoft Corp., Redmond, WA). This study used an approach requiring that an initial microbiological environmental sampling plan be based on the answers to a series of questions related to product hazards, processing risks and controls, and knowledge of appropriate microbiological sampling and testing protocols. Furthermore, the initial sampling plan can be related to the volume of product and size of the processing facility.

These sampling plans will provide the user with the total number of samples to collect each month for a pathogen (qualitative) and an indicator organism (quantitative). Furthermore, these sample quantities are distributed between three environmental sampling zones. Additional sampling guidance is provided.

The outputs of the initial sampling plan design spreadsheets were linked to another spreadsheet that can be used to record and summarize environmental sampling test results. Together these were customized to create an interactive tool that will suggest modifications to the current sampling plan based on the cumulative test results obtained over one month and three months. The development of these sampling plan design tools are described below.

## **A. Development of an Interactive Tool to Establish an Environmental Sampling Plan for an Infant Formula/ Infant Cereal Processor**

### **1. Initial sampling plan based on product and process food safety risk**

To setup a microbiological environmental sampling plan or to validate an existing sampling plan several factors must be considered that require some knowledge of how the product is produced, the microbial hazards that could be found in the raw ingredients or introduced in the process, the food safety controls used in the processing plant, the number and volume of products produced over time, and the appropriate qualitative or quantitative microbiological tests that are needed. Additionally, the sampling guidance does not assume that the user has conducted microbiological sampling and testing in the past or has knowledge of best practices for sampling or has knowledge of the previous level of environmental sampling for a processing plant.

A typical food processing plant has numerous locations or zones where an environmental sample could be collected. Each sampling zone may have a specific hygiene requirement and unacceptable test results from one or more samples within a zone may require a different response from the processor. Frequently, food processors categorize environmental samples as originating from one of these three zones:

- Zone 1 – includes in-process product and surfaces that can contact the product;
- Zone 2 – encompasses the areas directly adjacent to Zone 1 and includes all non-food contact or indirect contact areas in the processing plant;
- Zone 3 – sampled areas that are usually environmental (floors, ceilings walls) and typically not in the food processing rooms.

In the initial setup of the sampling plan at least one sample must be collected from each assigned zone location spread throughout the processing line.

## **2. Questionnaire to determine relative product/process risk:**

A questionnaire was designed to elicit responses on food safety procedures, processing conditions, and sampling and testing protocols for a processor of powdered infant formula or infant cereal. These are low-moisture foods that may be considered unsafe due to possible contamination by the microbial pathogens *Salmonella* spp. and *Cronobacter sakazakii*.

An initial list of 80 questions was sorted and ranked by their importance for affecting the relative food safety of the product and process. The final list of 22 questions are discussed below and presented in Figure 1. The process of developing the sampling monitoring plan was based on three sets of questions:

### **Step One: Food safety procedures**

The questions listed below ask if the user practices or is aware of some basic food safety related operations including Good Manufacturing Practices, monitoring of ingredients and employee training.

- Do you follow Good Manufacturing Practices (GMPs)?
- Do you have an environmental monitoring sampling plan (including sample locations, frequencies, types, sizes)?
- Do you know the source of your raw materials and ingredients?
- Do you maintain a certificate of analysis for your materials?

- Do you require specifications for all ingredients?
- Did / Will your staff receive food safety and GMP training this year?

### **Step Two: Processing hazards**

The questions listed below ask about procedures that may increase or decrease the relative microbial safety of the product. Also, the questions help ascertain if the user has knowledge of the potential hazards and food safety controls in their process and if they can provide the required measures to control the processing environment. As a control measure, the staff food safety awareness is vital to handle this type of food. The sources or frequency of contamination can be lowered or eliminated by following good manufacturing and hygienic practices.

- What is the physical condition of your process facility?
- Does your processing room have positive pressure?
- Do you use a HEPA filter in you air unit?
- What type of barrier between zones do you have in the process line?
- Are you knowledgeable of the type of hazards to your product?
- What is the risk of pathogen contamination to your product?
- Does the risk of your product to the consumer change after production?
- Is any source of pathogen contamination eliminated from the process line?

### **Step Three: Microbiological Sampling and Testing**

The questions listed below are used to verify the user's awareness of the zoning concept and current sampling plan, if any, for the environment and product.

Microbial sampling and testing is commonly used to validate and verify effective hazard control measures in place.

- Do you know the (pathogenic) microorganism of concern?
- Do you test product samples for pathogens?
- Do you test for indicator or non-pathogenic organisms?
- Is your staff trained to collect environment samples?
- Do you know how many environmental samples you take per lot or per day?
- Do you have a list of locations to collect environment samples from?
- How many environmental sampling zones do you have?
- Do you know the microbiological test specifications of your analyses?

### **3. Questionnaire to determine relative production volume/ plant size:**

Five additional questions were added to the questionnaire described above, to elicit responses for categorizing the production volume of a powdered infant formula or infant cereal processor. These questions asked about the daily operating hours of the plant, number of employees, annual production volume and annual sales volume.

Generally, food processors will collect more environmental samples in larger plants since there are more locations to sample and more places that pose a risk for product cross-contamination. These additional questions are listed below and in Figure 2.

- How many work shifts per day for production?
- How many production lines do you have in your facility?
- How many employees you have?
- What is your production volume per year (units or pounds)?

- What is your sales volume per year?

#### **4. Question response scoring**

For each question, four choices were provided. In many cases, the four choices include the responses of 1) “yes”; 2) “no”; 3) “usually”, “sometimes” or “not sure”; and 4) “don’t know” or “unknown”. For each question, the responses were assigned a value ranging from 1 to 5. A value of 5 reflected that the response could be related to improved food safety. A lower value implies that these responses are detrimental to food safety or do not enhance food safety. The response of “don’t know” or “unknown” was always scored as “1”. For the questions related to production volume and size of operations, the four choices for each question carried a number or a range of numbers. A value of 4 or 5 reflected that the selected response represented a larger size or volume plant, and an answer that reflected a relatively small volume plant resulted in a score of 1 or 2 (Figure 1 and 2).

Each of the concepts represented by the 27 questions was ranked as having a relatively high to low food safety impact or significance to the sampling plan. Multiplier factors were used for each ranked question depending on its importance to form the sampling plan. The multipliers were 10 (low), 20 (medium) and 30 (high). The higher the multiplier meant the question was relatively more important for food safety impact.

To determine the relative risk level of the product and process, the scores for each of the first 22 question responses can be multiplied by their food safety impact score (10, 20, or 30) and summed. The total score possible ranges from 420 to 2100.

Twenty combinations of question responses were used to determine appropriate total score ranges that would correspond to low, medium and high process risk (Table 1).

To determine the relative production volume of the processing plant, the scores for each of the final 5 question responses can be multiplied by their food safety impact score (20 or 30) and summed. The total score possible ranges from 220 to 600. Twenty combinations of question responses were used to determine the appropriate total score ranges that would correspond to low, medium and high production volume (Table 1).

## **5. Sampling plans based on process risk and production volume**

Nine basic sampling plans were created based on product/ process risk levels (low, medium or high) and production volume levels (low, medium or high). Each of these sampling plans will provide the user with the total number of samples to collect each month for a qualitative pathogen test (*Salmonella*, for example) and a quantitative indicator organism (Enterobacteriaceae, for example). For the infant formula/ infant cereal product and process used in this example, each environmental sample is tested for both a pathogen (*Salmonella*) and an indicator organism (Enterobacteriaceae (EB) count). Furthermore, these sample quantities are distributed between three environmental sampling zones (Table 2). The sampling plans take into consideration two main factors- sampling frequency and zone location. The sampling frequency represents how often samples must be taken from each zone. Each zone should be tested for any pathogen contamination, but the number of samples from each zone may vary.

To illustrate further, the number of sample collected for the process line is determine by the production volume, employee number and number of processing line. The bigger the production volume the more samples would be collected. Another factor that might increase the sampling number includes how many shifts would the operation run per day. Due to the increase of the operational shifts per day, more samples must be attained to validate the collected data over a specific period of time (Figure 2).

## **B. Development of a Spreadsheet Tool to Record and Summarize Environmental Sample Test Results for an Infant Formula/ Infant Cereal Processor**

### **1. Recording environmental sample test results**

A spreadsheet template for recording and evaluating environmental sampling data was developed based on the format described by Eifert and Arritt (2002). This template provides a format for recording environmental sample identification information including time of collection (day, date, shift), plant area location, sample location, analytical test (qualitative or quantitative) and test result. For the current project, a data set of environmental sample test results was constructed for test purposes and analyzed using the "PivotTable" feature in Microsoft® Excel 2010 (Microsoft Corp., Redmond, WA). A PivotTable is interactive tables that quickly summarizes, or cross-tabulates, large amounts of data, including user-selected subsets of the data. The user can rotate the rows and columns to see different summaries of the source data, filter the data by displaying different pages, display the details for areas of interest, and ultimately chart the PivotTable data.

The data set constructed for this example contained 360 line entries with test results for quantitative (*Enterobacteriaceae*) tests and qualitative (*Salmonella*) tests. A



portion of the data table, shown in Table 3, includes the following "Fields" and accompanying data ranges.

- Date sample taken:
- Date sample analysis on:
- Environmental Zone (1, 2, or 3)
- Sample site (within a zone) code or abbreviation
- EB count test result (CFU/mL)
- Salmonella test result: "1" = positive, "0" = negative, blank = no test
- EB count test decision: sample fails when counts exceed 10/mL from zone 1, 100/mL from zone 2, and 500/mL from zone 3

While our example data set required data input directly into the cells of the spreadsheet, the environmental sampling tool was designed so that users will enter sample information into a separate dialog box for each sample. That information will then be transferred to the cumulative record of sample information. Examples: Date of analysis used to validate the compliance with any policy implemented in the manufacturer lab. The date of analysis can also be used to verify if the sample had sufficient time to be analyzed. The "Date sample taken" inputs can verify if the sampling plan frequency is sufficient to be conducted in a timely manner.

## **2. Sample Test Result Summaries**

Summaries of the sample description and test result data can be reported with Pivot Tables or Pivot Charts to show trends. Numerous combinations of data variables and table and chart formats are possible. Moreover, the collected data can be

represented in trends to illustrate the need to increase or decrease the control level on the processing environment. Pivot Tables of the test result summaries can be used to present test results of a specific zone, period of time or pathogens of concern and indicator microbe. Evaluating the test result summaries can enhance the decisions to be made to improve the controls implemented on site. Correlation between indicator and pathogen analytical test results, over time, obtained from processing environments can be investigated for trends, but comparison for individual samples is usually not recommended. Comparing test results between or within zones can illustrate the source of any contamination. The sampling tool will be designed to display a minimum number of data summaries to reduce confusion. To create additional data summaries, users will be able to export the sample information data for further manipulations.

### **C. Sampling Plan Modification based on Monthly Evaluations of Test Results**

Evaluating the data collection over a given time period can support the decision to modify the initial sampling plans. Test result evaluation can determine the need to increase or decrease the sampling number obtained from the process line environment. The sampling tool will be designed to evaluate test results over a specific time period. When this function the program will then inform the user that the sampling plan may need to be changed in the next time period (the following month, for example). The guidelines included in the sampling tool will specify the general changes in the sampling required that will cause an increase or decrease in the number of samples collected in each zone in the following month. For both pathogenic and indicator test results over time, four specific guidelines will be used to determine the need to change the initial sampling

plan. If one or more of the specified guidelines were compromised the sampling plan will change as per the described guidelines.

In the guidelines, two time periods were specified, one- and three- months of data collection to direct the change in the initial sampling plan. After one month of data collection if the initial sampling plan test result complied with the given guideline the same sampling plan shall continue to the next month cycle. Unless one or more of the guidelines are compromised the initial sampling plan shall shift to a higher strict sampling plan, for a specified time described in the guideline. If the test results obtained from the environmental samples are below the limit for unacceptable results, then the sampling will return to the lower sampling plan. If the test result obtained from the environment samples were positive the higher sampling plan shall continue until the process environment comes under control. If the data collected from the processing environment test result were negative for a consecutive three month, the sampling plan shall decrease to 10% of the total initial sampling plan size. The new reduced sampling plan will be evaluated for one month. If the new decreased sampling plan leads to results that are below the fail limit over a consecutive three month cycle, then the sample plan shall decrease for an additional 10%. An overall reduction of 20% from the initial sampling plan is the maximum sampling plan decrease.

The number of samples required for each of the 9 sampling plans at sampling state 0 (initial plan) and sampling plan state 1, 2, 3, and 4 are shown in Tables 4, 5, 6, and 7, respectively. The sampling plan tool will alert the user when one of these four situations occurs that will result in a change in the monthly sample plan by zone. For sampling state 1 and 2, the number of samples in the next plan will be higher if the

number of “fail” samples in a month exceeds the limit for *Salmonella* or Enterobacteriaceae, respectively. For sample state 3, exceeding the fail limit 3 months in a row will result in this new sampling plan. If the number of samples that fail in a month is below the limit, for 3 consecutive months, then the number of samples will be reduced by 10% the following month for sample state 4.

## RESULTS

The “Sampling Test Improvement Workbook” developed for this thesis research project is available as an electronic file (\*.xlsm) in Microsoft Excel. The inputs and features of this software program are further described below. Each input can be used to improve the sampling plan tool. The inputs integrate with each other, to provide data summaries and sampling plan recommendations. The data can be presented as needed to support sampling decisions made to improve or maintain the hygienic condition in the process line environment.

Selected screenshots of the environmental sampling tool software program are included in Figures 3 – 7. The parameters of the current sample plan, with the number of samples to collect at each site, are shown in Figure 3. An example of a list of sample sites and zones is shown in Figure 4. Sample location descriptions can be abbreviated with a two-character code to facilitate data entry. An example of two Pivot Table summaries for Enterobacteriaceae “EB” or *Salmonella* “S” test results by zone and site is in Figure 5. Figure 6 displays the sample date, site and test result window for the software user. Finally, in Figure 7, the initial sample plan software interface is displayed.

After defining the inputs requested by the sampling plan program, an initial sampling plan will be selected which can be modified based on sample test results. Unsatisfactory test results may change the sampling plan state and sample number. The increase and decrease of the sampling plan shall change upon the test result entered to the software program. The minimum number of samples is one per processing location.

For the environmental sampling monitoring tool a set of questions was developed to focus on the product and process food safety risks. The level of hygienic control in the process line can be determined from the risk of the product, product exposure and strictness of the sampling plan. Another set of questions was used to estimate the relative production volume for the starting point of the sampling plan. The bigger the volume the more control must be implemented on the level of hygienic control.

For each question and set of responses, point scores were assigned to guide the initial sampling plan as per the process needs. A higher score determines the food manufacturers' awareness of the process requirements and controls implemented on site. The higher the score the less strict and fewer samples the sampling plan will be, with consideration of the sample location which must be sampled at least once per week. The formation of nine sampling plans is included in the program software. Upon the answered questions and scores attained, a specific sampling plan would be assigned to each process facility. A recommendation is given to start from the higher level of the hygienic control to a lowest level of control. Samples to collect each month from any group would be categorized for each zone 1, 2, 3 in order of 50, 30, and 20% of the total.

For each sample collected from an individual location both pathogen (qualitative) and indicator (quantitative) tests are recommended. The collected data result from both tests shall be analyzed for further assessment of the food manufacturer process line hygienic condition and decision shall be made to increase or decrease the sampling size as per the advised sampling plan.

A specific guideline for each state of sample collection is described in the initial sampling monitoring plan. Guidelines illustrate the mechanism of the sample collection size and location. Moreover, unsatisfactory test results from any collected sample shall trigger a change in the sampling number, location and frequency. Guidelines provide the limits and specification to change the sampling plan. Guidelines are subject to change as needed by the food manufacturer. The rule of increase and decrease the sampling plan size is described in the guidelines. Limits of accept or reject any test result is described as well in the guidelines. The start and end of the sampling plan cycle is written in the guidelines as well.

Each sampling plan has three hygienic levels of control which have been designated as a “State”. Any change to the sampling plan shall be between the same set of hygienic level of control. The higher the state the more strict the sampling requirement would be (higher number of samples to collect). A guideline is provided to describe the change between sampling states.

## DISCUSSION

Development of an appropriate environmental sampling plan is essential to determine the readiness of the production facility to produce a potentially hazardous food product. This environmental sampling tool provides a method of creating a sampling plan with only basic knowledge of the product, process, hazards, and volume of the production. This basic knowledge can be driven from experience or process needs. The data outcomes from the initial sampling plan can be analyzed and decision can be made from it. The initial sampling plan must comply with the process needs as well as the regulatory authority.

The initial question set provides a way to initiate the process of determining the appropriate number of samples to collect for initiating a new sampling plan, as well as to clarify what are the main food safety concerns in the process line. These questions and response choices were designed so that someone with incomplete knowledge of the manufacturing operation could design a sampling plan, and so that a facility could consider using this tool even if they already were collecting microbiological environmental samples.

Questions can be added that can elicit specific information about the product and process or can be modified to meet the manufacturer needs and concerns. The questions and question responses can be re-weighted as needed by the manufacturer or alternate responses could be created. Moreover, the questions can be presented in an alternate order to meet the manufacturer need starting from the end to beginning of the process line. The question set can also guide the manufacturer for how many



samples they have to collect from the process line after specifying the number of locations in their process. Furthermore, these questions could be used to determine if procedures have been established that are specific to a Hazard Analysis Critical Control Program (HACCP). Having such control procedures in place is a significant way to reduce or eliminate the food safety hazard that threatens your product. Adapting HACCP is important to ensure that the product is produced under well-established measures. Taking into consideration the pathogen of concern and the detection method, would assure the awareness.

General questions on the production volume, sales volume, number of employees, and the length of daily operations, were used to determine three levels of total sampling plan size needed to be collected from the process line environment. Sampling frequency can be driven from the question set as for how many shifts the operation is running. As stated previously, all the questions used can be customized as per the manufacturer's needs and compliance with the regulatory authority. Having a schedule sampling plan is very important to avoid any gap in the sampling plan. A proactive sampling plan is important to ensure that the food is delivered under a safe environment.

The environmental sampling tool utilizes the zoning concept to define and organize sampling sites in a process facility. Complying with the zoning concept is significant, to ensure that each individual zone has been tested for any sign of contamination. Testing each zone and evaluating the result will assess the hygienic condition of the process line. The proportion of environmental samples to collect in each of the three environmental zones, suggested by the program, emphasizes a higher

number of samples in zone 1, and the fewest in zone 3. Description of each sample location is written in the sampling plan to refine the sampling location. The sample proportion should be verified by a food manufacturer. The estimated minimum number of samples taken from the process line must be at least one sample from each zone location for a specific time period. Users of this program may want to alter the starting percentages, but those changes would require access to macros and Visual Basic code used to develop the sampling tool.

The number of environmental samples which must be collected from the processing area is very crucial. Not only must the processor ensure that the food product is produced under hygienic condition, they may also need to comply with a regulatory authority which could require that a minimum number of environmental samples are collected and tested. Food manufacturers may want to pool or combine samples within a zone or from a site prior to conducting a microbial analysis. This procedure can reduce the number of samples taken from the process line, but it must be verified and validated by the food manufacturer. Traceability of each sampled location is a must, to ensure the accuracy of the environment sampling monitoring plan.

For data entry, a guideline is provided to support the decision of any changes to the sampling plan. Guidance is given to determine when the sampling plan must be changed and when the new sampling plan shall start, as well as if any unsatisfactory result is entered what corrective action should be made. In the sampling plan a specific sample number derivative from the number of sampling location.

Test result data can be easily summarized to determine if actions are needed, if the plan needs modification, or to evaluate trends. Sample test data records and test

result summaries could be modified to include: sample results on a weekly basis, mean quantitative test results, sample size or area specifications, and randomized selection of sample sites within a zone, for examples.

Trends observed in the sample test results can indicate the level of hygiene of the process line or facility. This information can illustrate the need to increase or decrease the control on the processing environment by adjusting the number of samples collected from the process line. Additional measures can be taken to improve and verify the hygienic condition of the process line environment on an immediate or long-term basis as needed. The availability to the data collected over a year or more can support the decision made to improve or change the sampling plan or other food safety programs.

The microbiological environmental sampling plan tool can be modified for other food products besides low-moisture foods or milk-based products. Adaptation to other food products and processes would require some knowledge of the food process, the finished product, the intended user of the product, the microorganisms of concern, the recommended tests for these microorganisms, the likelihood of product or process contamination, and regulatory testing requirements. While this approach for creating and modifying an environmental sampling plan can improve the hygienic condition of the process line environment and enhance food safety, a food processor may still need to conduct appropriate microbial testing on finished products.

## SUMMARY

Complying with the GHP in process lines is essential to prove that the food is produce under a sanitary condition. The hygienic condition of the processing environment is conceder a critical factor to food safety. Collecting samples from the processing environment indicate the hygienic condition of the process line. Developing a suitable sampling monitoring plan is crucial to control the source of contamination.

The developed sampling plan can support the decision made to increase or decrease the environment sampling number. Evaluating the data collected for the test result obtained from pathogenic and indicator analysis can illustrate the need to change or modify the initial sampling plan. Features included in the software program to assess the need to change that sampling plan accordingly. Pivot tables and trend analysis represent the data collected of a specific need. Improving and controlling the hygienic condition of a process should be a concern of a food manufacturer. Additionally, using such a tool can illustrate the need to take major corrective actions, if needed.





**Figure 3.** Current sampling plan view describes the sample plan category code, risk level, current guideline state, production volume level, data entry and data report view.

# Current Plan

Cycle Start: 4/20/2013  
Date: 4/26/2013

**Sampling Plan:** 2C      medium      **Risk:** high

**Current State:** 0

Enter new data

View Reports

Zone	Site	Taken:	Goal:	Remaining:	Completed:
1	1a	2	9	7	✗
	1b	2	9	7	✗
	1c	2	9	7	✗
	1d	2	9	7	✗
	1e	3	9	6	✗
	1f	1	9	8	✗
	1g	1	9	8	✗
	1h	1	9	8	✗
	1i	1	9	8	✗
	1j	1	9	8	✗
	2	2a	2	7	5
2b		2	7	5	✗
2c		1	7	6	✗
2d		1	7	6	✗
2e		1	7	6	✗
2f		1	7	6	✗
2g		1	7	6	✗
2h		0	7	7	✗
3	3a	2	8	6	✗
	3b	1	8	7	✗
	3c	1	8	7	✗
	3d	1	8	7	✗
	3e	1	8	7	✗
<b>Grand Total</b>		31	186	155	✗

**Figure 4.** Sample site view describes site code, zone, sample description or location.

Site	Zone	Description
1a	1	Conveyor Belt 1
1b	1	Conveyor Belt 2
1c	1	Conveyor Belt 3
1d	1	Processing Machine 1
1e	1	Processing Machine 2
1f	1	Main area floor (left)
1g	1	Main area floor (right)
1h	1	Main area ceiling
1i	1	Main area west wall
1j	1	Main area east wall
2a	2	Hallway 1
2b	2	hallway 2
2c	2	hallway 3
2d	2	Facility North wall
2e	2	Facility South wall
2f	2	General office 1
2g	2	General Office 2
3a	3	Facility Entrance
3b	3	Emergency Entrance 1
3c	3	Emergency entrance 2
3d	3	Shipping Dock 1
3e	3	Shipping Dock 2
2h	2	Random Area 1

These are your sampling sites. Please remember to **ALWAYS** assign a site to zone "1", "2", or "3".

Once you have finished editing sites, press the button below.

**Edits Complete**



**Figure 5.** Summary report pivot table for both enterobacteriaceae “EB” count and *Salmonella* “S” test result, including zone, total number of samples and site location.

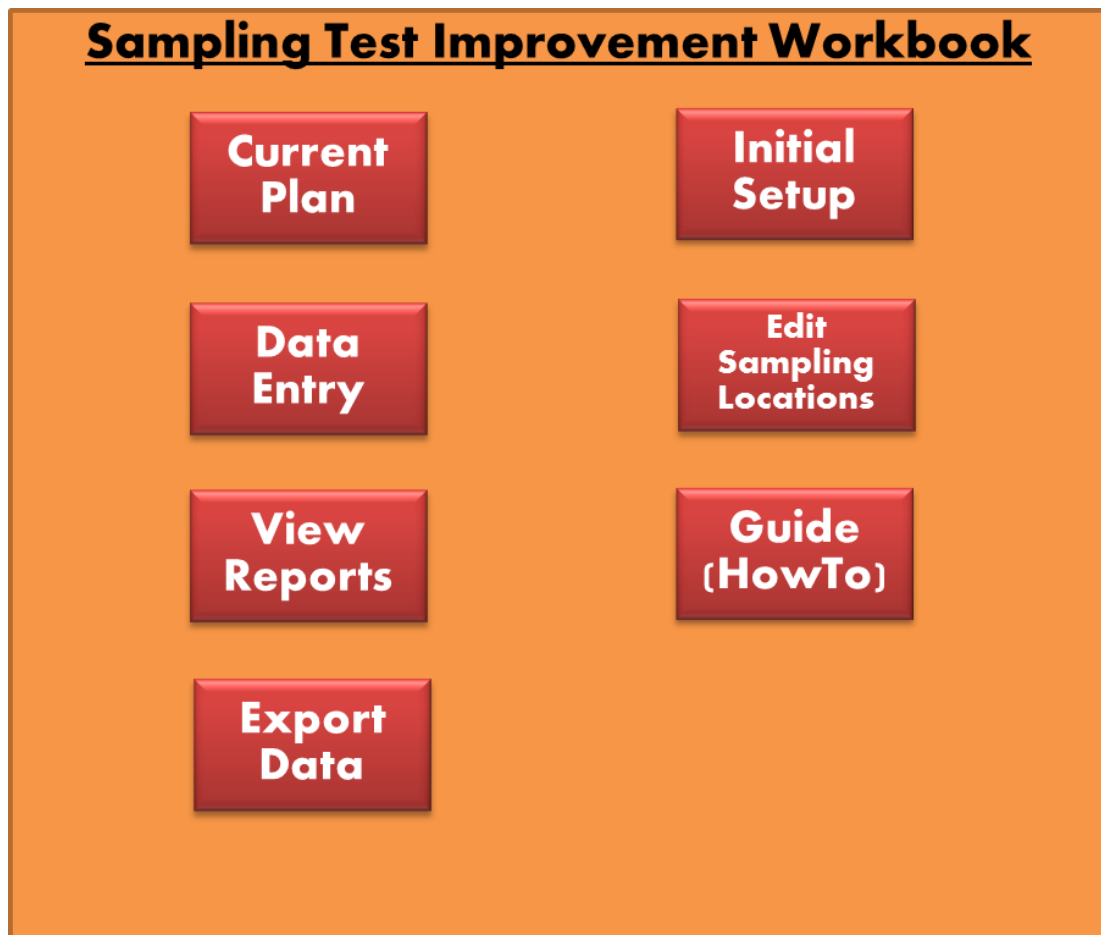
Current Cycle Results					S-Test Results					
Current	1	EB_pass	Fail	Pass	Total	Current	1	S_Test	Pass	Total
Total #	Zone	Site	Fail	Pass	Total Samples	Total #	Zone	Site	Pass	Total Samples
	1.1a				2		1.1a			2
	1b	1			1		1b			2
	1c				2		1c			2
	1d	1			1		1d			2
	1e				3		1e			3
	1f				1		1f			1
	1g				1		1g			1
	1h				1		1h			1
	1i				1		1i			1
	1j				1		1j			1
<b>1 Total</b>			<b>2</b>		<b>14</b>	<b>1 Total</b>			<b>16</b>	<b>16</b>
	2.2a				2		2.2a			2
	2b				2		2b			2
	2c				1		2c			1
	2d				1		2d			1
	2e				1		2e			1
	2f				1		2f			1
	2g				1		2g			1
<b>2 Total</b>					<b>9</b>	<b>2 Total</b>			<b>9</b>	<b>9</b>
	3.3a				2		3.3a			2
	3b				1		3b			1
	3c				1		3c			1
	3d				1		3d			1
	3e				1		3e			1
<b>3 Total</b>					<b>6</b>	<b>3 Total</b>			<b>6</b>	<b>6</b>
<b>Total Samples</b>			<b>2</b>		<b>29</b>	<b>Total Samples</b>			<b>31</b>	<b>31</b>

Main Menu

Current Plan



**Figure 7.** Initial sample plan software interface.



**Table 1.** Summary of question response scores required for each of the 9 possible starting environmental sampling plans.

<b>Volume Question Response Scores Sum</b>	<b>Plant Volume</b>	<b>Product/ Process Risk Question Response Scores Sum</b>	<b>Product/ process risk</b>	<b>Initial Sample Plan Code</b>
<b>&lt;300</b>	<b>low</b>	<b>&gt;1500</b>	<b>low</b>	<b>1A</b>
<b>&lt;300</b>	<b>low</b>	<b>1000-1499</b>	<b>medium</b>	<b>1B</b>
<b>&lt;300</b>	<b>low</b>	<b>&lt;1000</b>	<b>high</b>	<b>1C</b>
<b>300-450</b>	<b>medium</b>	<b>&gt;1500</b>	<b>low</b>	<b>2A</b>
<b>300-450</b>	<b>medium</b>	<b>1000-1499</b>	<b>medium</b>	<b>2B</b>
<b>300-450</b>	<b>medium</b>	<b>&lt;1000</b>	<b>high</b>	<b>2C</b>
<b>&gt;450</b>	<b>high</b>	<b>&gt;1500</b>	<b>low</b>	<b>3A</b>
<b>&gt;450</b>	<b>high</b>	<b>1000-1499</b>	<b>medium</b>	<b>3B</b>
<b>&gt;450</b>	<b>high</b>	<b>&lt;1000</b>	<b>high</b>	<b>3C</b>

**Table 2.** Summary of total environmental samples required, and maximum number of unacceptable (failed) samples, for each of the 9 possible starting environmental sampling plans.

Initial Sample Plan	Plant Volume	Product/ process risk	Environmental samples / month				Sample fail limit per month	
			Total	zone 1 (50%)	zone 2 (30%)	zone 3 (20%)	"S"	"EB"
1A	low	low	30	15	9	6	1	10
1B	low	medium	60	30	18	12	1	20
1C	low	high	90	45	27	18	1	30
2A	medium	low	60	30	18	12	1	30
2B	medium	medium	120	60	36	24	1	50
2C	medium	high	180	90	54	36	1	70
3A	high	low	90	45	27	18	1	40
3B	high	medium	180	90	54	36	1	70
3C	high	high	270	135	81	54	1	120

All samples can be tested for both *Salmonella* "S" (qualitative) and Enterobacteriaceae "EB" (quantitative)

Monthly sample total should be divided so that an equivalent number is collected each week.

Number of sample sites per zone per week should be at least 3 unless fewer samples are required

**Table 3.** Data collection report view includes date sample taken, date sample analyzed, sampling zone, sample site code, enterobacteriaceae “EB” test result, *Salmonella* “S” test result, and sample test result validation (Pass/Fail).

DateTaken	DateAnalyzed	Zone	Site Code	EB_Test	S_Test	EB_pass
3/1/2013	3/2/2013	1	1a	0	0	Pass
3/2/2013	3/2/2013	1	1a	0	0	Pass
3/3/2013	3/4/2013	1	1b	20	0	Fail
3/3/2013	3/4/2013	1	1b	0	0	Pass
3/3/2013	3/4/2013	1	1c	12	0	Fail
3/3/2013	3/4/2013	1	1c	0	0	Pass
3/3/2013	3/4/2013	1	1d	4	0	Pass
3/3/2013	3/4/2013	1	1d	12	0	Fail
3/4/2013	3/4/2013	1	1e	33	0	Fail
3/4/2013	3/4/2013	1	1e	100	0	Fail
3/4/2013	3/4/2013	1	1f	0	0	Pass
3/4/2013	3/4/2013	1	1g	0	0	Pass
3/4/2013	3/4/2013	1	1h	0	0	Pass
3/4/2013	3/4/2013	1	1i	120	0	Fail
3/4/2013	3/4/2013	1	1j	0	0	Pass
3/5/2013	3/6/2013	2	2a	0	0	Pass
3/5/2013	3/6/2013	2	2a	0	0	Pass
3/5/2013	3/6/2013	2	2b	30	0	Pass
3/5/2013	3/6/2013	2	2b	0	0	Pass
3/5/2013	3/6/2013	2	2c	90	0	Pass
3/6/2013	3/6/2013	2	2d	0	0	Pass
3/6/2013	3/6/2013	2	2e	0	0	Pass

**Table 4.** Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 1 with guideline for adjusting sample plan totals by zone (“if # of failed *Salmonella* samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, until you receive a negative result for 2 weeks”).

Initial Sample Plan	Plant Volume	Product/ process risk	Salmonella samples per month			Total Salmonella sample fail limit per month	State 1				
			zone 1 (50%)	zone 2 (30%)	zone 3 (20%)		z1	z2	z3		
			Total				Total				
1A	low	low	30	15	9	6	1	52	40	27	if # of failed samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, Till you receive a negative result for 2 week.
1B	low	medium	60	30	18	12	1	52	40	27	
1C	low	high	90	45	27	18	1	52	40	27	
2A	medium	low	60	30	18	12	1	90	54	36	if # of failed samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, Till you receive a negative result for 2 week.
2B	medium	medium	120	60	36	24	1	90	54	36	
2C	medium	high	180	90	54	36	1	90	54	36	
3A	high	low	90	45	27	18	1	135	81	54	if # of failed samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, Till you receive a negative result for 2 week.
3B	high	medium	180	90	54	36	1	135	81	54	
3C	high	high	270	135	81	54	1	135	81	54	

**Table 5.** Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 2 with guideline for adjusting sample plan totals by zone (“If # failed samples in a month > than limit, then increase EB samples by 2X each zone for 1 month until you receive under norm results”).

Initial Sample Plan	Plant Volume	Product/ process risk	EB samples per month				Total EB sample fail limit per month from all zones	State 2		
			Total	zone 1 (50%)	zone 2 (30%)	zone 3 (20%)		z1	z2	z3
1A	low	low	30	15	9	6	10	30	18	12
1B	low	medium	60	30	18	12	20	60	36	24
1C	low	high	90	45	27	18	30	90	54	36
2A	medium	low	60	30	18	12	30	60	36	24
2B	medium	medium	120	60	36	24	50	120	72	48
2C	medium	high	180	90	54	36	70	180	108	72
3A	high	low	90	45	27	18	40	90	54	36
3B	high	medium	180	90	54	36	70	180	108	72
3C	high	high	270	135	81	54	120	270	162	108



**Table 6.** Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 3 with guideline for adjusting sample plan totals by zone (“if # of failed samples > limit for 3 consecutive months, then shift to higher risk sampling plan (low --> med., or med. --> high) for 1 months till you receive under norm results”).

Initial Sample Plan	Plant Volume	Product/ process risk	EB samples per month				Total EB sample fail limit per month from all zones	State 3				
			Total	zone 1 (50%)	zone 2 (30%)	zone 3 (20%)		z1	z2	z3		
				30	15	9					6	
1A	low	low	60	30	18	12	10					
1B	low	medium	90	45	27	18	20	30	18	12		
1C	low	high	120	60	36	24	30	45	27	18		
2A	medium	low	180	90	54	36	30	60	36	24		
2B	medium	medium	270	135	81	54	40	90	54	36		
2C	medium	high	360	180	108	72	50	120	72	48		
3A	high	low	450	225	135	90	70	135	81	54		
3B	high	medium	540	270	162	108	80	162	97	65		
3C	high	high	720	360	216	144	120	216	129	86		

**Table 7.** Number of samples required for each of 9 sampling plans at sampling state 0 (initial) and state 4 with guideline for adjusting sample plan totals by zone (“if # of failed samples < limit for 3 consecutive months, then you may reduce total # of sample analyses by 10% (by pooling samples) if you have a good history of your results”).

Initial Sample Plan	Plant Volume	Product/process risk	EB samples per month				Total EB sample fail limit per month from all zones	if # of failed samples < limit for 3 consecutive months, then you may reduce total # of sample analyses by 10% (by pooling samples) if you have a good history of your results		
			Total	zone 1	zone 2	zone 3				
				(50%)	(30%)	(20%)				
1A	low	low	30	15	9	6	10	z1	z2	z3
1B	low	medium	60	30	18	12	20	14	8	5
1C	low	high	90	45	27	18	30	27	16	11
2A	medium	low	60	30	18	12	30	41	24	16
2B	medium	medium	120	60	36	24	50	27	16	11
2C	medium	high	180	90	54	36	70	54	32	22
3A	high	low	90	45	27	18	40	81	49	32
3B	high	medium	180	90	54	36	70	41	24	16
3C	high	high	270	135	81	54	120	81	49	32
								122	73	49

## REFERENCES

- Cordier, J.-L. 2008. Production of powdered infant formula and microbiological control measures, pp. 145-185. In J. M. Farber, and S. Forsythe (eds.). *Enterobacter sakazakii*. ASM Press. Washington, DC.
- Dahms, S. 2003. Microbiological sampling plans –Statistical aspects. In Mitt. Lebensm. Hyg. 95 (1): 32-44.
- EFSA (European Food Safety Authority). 2007. Opinion of the Scientific Panel on Biological Hazards (BIOHAZ) on the request for review of the opinion on microbiological risks in infant formulae and follow-on formulae with regard to Enterobacteriaceae as indicators. Adopted on January 24, 2007. The EFSA Journal. 444:1-14.
- Eifert, J.D. and Arritt, F.M. 2002. Evaluating environmental sampling data and sampling plans. Dairy, Food and Environ. Sanit. 22(5): 333-339.
- Environmental Protection Agency (EPA). December, 2002. Guidance on Choosing a Sampling Design for Environmental Data Collection. Retrieved, May 17 2011, from: [www.epa.gov/QUALITY/qs-docs/g5s-final.pdf](http://www.epa.gov/QUALITY/qs-docs/g5s-final.pdf)
- Farmer, J. J., M. A. Asbury, F. W. Hickman, and D. J. Brenner. 1980. *Enterobacter sakazakii*—a new species of Enterobacteriaceae isolated from clinical specimens. Int. J. Syst. Bacteriol. 30:569–584.
- Grocery Manufacturers Association (GMA). 2009. Control of *Salmonella* In low-moisture foods, February 4, 2009 (Minor corrections March 16, 2009). Washington, DC: Grocery Manufacturers Association.

- Institute of Food Technologists (IFT). 2004. Scientific Status Summary: Bacteria Associated with Foodborne Diseases. Retrieved, April 03 2012, from <http://www.ift.org/knowledge-center/read-ift-publications/science-reports/scientific-status-summaries/bacteria-associated-with-foodborne-diseases.aspx>
- International Commission on Microbiological Specifications for Foods (ICMSF). 2011. Milk and Dairy products, Ch. 23 In: Microorganisms in Foods 8: Use of Data for Assessing Process Control and Product Acceptance. Springer: New York City, New York.
- International Commission on Microbiological Specifications for Foods (ICMSF). 2002. Sampling to Assess Control of the Environment, Ch. 11, In: Microorganisms in Foods 7: Microbiological testing in food safety management. Kluwer Academic Plenum Publishers: New York City, New York.
- International Commission on Microbiological Specifications for Foods (ICMSF). 1986. Microorganisms in Foods 2: Sampling for Microbiological Analysis: Principles and Specific Applications. University of Toronto Press: Toronto.
- Jay LS, Davos D, Dundas M, Frankish E, Lightfoot D. 2003. *Salmonella*. In: AD Hocking (eds). Foodborne Microorganisms of Public Health Significance. Sixth edition. Waterloo, NSW: Australian Institute of Food Science and Technology Incorporated, NSW Branch, Food Biology Group.
- Legan, D., & Vandeven, M. H. 2003. Sampling techniques. In T. McMeekin (Ed.), Detecting Pathogens in Food (pp. 20-51). Cambridge, London: Woodhead Publishing Limited

- Marriott, N. G. and Gravani, R. B. 2006. Principles of Food Sanitation: Food Contamination Sources (pp. 78). New York, New York: Springer Science + Business Media, Inc.
- New Zealand Food Safety Authority (NZFSA), October, 2010. Institute of Environmental Science and Research Limited (ESR): Risk profile: Salmonella (non typhoidal) in cereal grains.
- NIST. 2012. Challenges in Microbial Sampling in the Indoor Environment – Workshop Summary Report. National Institute of Standards and Technology (NIST) Technical Note 1737. Retrieved, Jan 18 2012, from: [www.nist.gov/](http://www.nist.gov/)
- Norberg, S., Stanton, C., Ross, R. P., Hill, C., Fitzgerald, G., & Cotter, P. 2011. *Cronobacter* spp. in powdered infant formula. Journal of Food Protection, 75(3), 607-620.
- Scallan, E., Hoekstra, R.M., Angulo, F.J., Tauxe, R.V., Widdowson, M.A., Roy, S.L., Jones, J.L., Griffin, P.M. 2011. Foodborne illness acquired in the United States--major pathogens. Emerg. Infect. Dis. 17: 7-15.
- Sutton, S. 2010. The environmental monitoring program in a GMP environment. Journal of GXP Compliance 14(3): 22-30.
- Tompkin, B. 2004. Environmental sampling – A tool to verify the effectiveness of preventive hygiene measures. In Mitt. Lebensm. Hyg. 95 (1): 45–51.
- U.S. Food and Drug Administration (FDA). 2012 Bad Bug Book (second edition): Foodborne Pathogenic Microorganisms and Natural Toxins Handbook. *Salmonella* spp. Retrieved, April 26 2012, from:

<http://www.fda.gov/downloads/Food/FoodSafety/FoodborneIllness/FoodborneIllnessFoodbornePathogensNaturalToxins/BadBugBook/UCM297627.pdf>

USDA FSIS. 2013. Intensified verification testing (ivt) protocol for sampling of product, food contact surfaces, and environmental surfaces for *Listeria monocytogenes* (lm) or *Salmonella* spp. U.S. Department of Agriculture, Food Safety and Inspection Service, Directive 10300.1. Retrieved, Feb10 2012, from: [www.fsis.usda.gov/OPPDE/rdad/FSISDirectives/10300.1.pdf](http://www.fsis.usda.gov/OPPDE/rdad/FSISDirectives/10300.1.pdf)

World Health Organization (WHO). 2007. Safe preparation, storage and handling of powdered infant formula guidelines. Switzerland: World Health Organization. Retrieved, Mar 17 2011, from: [http://www.who.int/entity/foodsafety/publications/micro/pif\\_guidelines.pdf](http://www.who.int/entity/foodsafety/publications/micro/pif_guidelines.pdf)

# Sampling Test Improvement Workbook

**Current  
Plan**

**Initial  
Setup**

**Data  
Entry**

**Edit  
Sampling  
Locations**

**View  
Reports**

**Guide  
(HowTo)**

**Export  
Data**

Developed by Hassan Masri and Ivan Volonsevich under guidance of Joseph D. Eifert  
Virginia Tech

# FS Procedures Questions

Do you follow Good Manufacturing Practices (GMPs)?

Yes

No

Usually

Don't Know

Do you have an environmental monitoring sampling plan (including sample locations, frequencies, types, sizes)?

Yes

No

Partially

Don't Know

Do you know the source of your raw materials and ingredients?

Yes

No

Sometimes

Don't Know

**Thanks for trying out the tool!**

To begin, please answer the questions listed here. Just scroll down until you answer all of the questions, and click "next" to see the next category of





Do you maintain a certificate of analysis for your materials?

- Yes
- No
- Sometimes
- Don't Know

Do you require specifications for all ingredients?

- Yes
- No
- Usually
- sometimes

Did / Will your staff receive food safety and GMP training this year?

- Yes
- No
- some employees
- only when hired



# Process Questions

What is the physical condition of your process facility?

- Very Good
- Good
- Fair
- unknown

Does your processing room have positive pressure?

- Yes
- No
- Usually
- unknown

\* Do you use a HEPA filter in you air unit?

- Yes
  - No
  - sometimes
  - unknown
-

What type of barrier between zones do you have in the process line?

- Physical
- Virtual
- both
- Don't Know

Are you knowledgeable of the type of hazards to your product?

- Yes
- No
- probably
- Don't Know

What is the risk of pathogen contamination to your product?

- High
- Medium
- Low
- not sure

Does the risk of your product to the consumer change after production?

- increased
- no change
- reduced
- not sure

Is any source of pathogen contamination eliminated from the process line?

- Yes
- No
- Usually
- not sure



Do you have a list of locations to collect environment samples from?

- Yes
- No
- not sure
- Don't Know

How many environmental sampling zones do you have?

- one
- two
- three or four
- Don't Know

Do you know the microbiological test specifications of your analyses?

- Yes
- No
- Somewhat
- Don't Know



**Next**

# Sampling and Testing Questions

\* Do you know the (pathogenic) microorganism of concern?

- Yes
- No
- not sure
- Don't Know

\* Do you know the (pathogenic) microorganism of concern?

- Yes
- No
- not sure
- Don't Know

Do you test product samples for pathogens?

- Yes
- No
- sometimes
- Don't Know

Do you test for indicator or non-pathogenic organisms?

- Yes
  - No
  - Sometimes
  - Don't Know
-

Is your staff trained to collect environment samples?

- Yes
- No
- Somewhat
- Don't Know

Do you know how many environmental samples do you take per lot or per day?

- Yes
- No
- not sure
- Don't Know

# Production Volume Questions

How many work shifts per day for production?

- one
- two
- one or two
- three

How many production line do you have in your facility?

- one
- two
- more than two
- not sure

How many employees you have?

- >500
  - 100-500
  - 10-100
  - <10
-



What is your production volume per year (units or pounds)?

- >1 million
- 100,000 - 1 million
- <100,000
- not sure

What is your sales volume per year?

- >\$ 1 million
- \$ 100,000 - 1 million
- < \$100,000
- not sure

**Finishe**

Based on our analysis of your answers, we have concluded that this is the risk level representative of your process.

risk:

**high**

Also, we have concluded that this is representative of your volume level.

volume:

**medium**

Based on these two factors, this is the suggested initial sampling plan.

Plan:

**2C**

The number represents volume. 1 is the lowest, 3 is highest. The letter represents

**Now you should enter your**

**Edit  
Sampling  
Locations**

Site	Zone	Description
1a	1	Conveyor Belt 1
1b	1	Conveyor Belt 2
1c	1	Conveyor Belt 3
1d	1	Processing Machine 1
1e	1	Processing Machine 2
1f	1	Main area floor (left)
1g	1	Main area floor (right)
1h	1	Main area ceiling
1i	1	Main area west wall
1j	1	Main area east wall
2a	2	Hallway 1
2b	2	hallway 2
2c	2	hallway 3
2d	2	Facility North wall
2e	2	Facility South wall
2f	2	General office 1
2g	2	General Office 2
3a	3	Facility Entrance
3b	3	Emergency Entrance 1
3c	3	Emergency entrance 2
3d	3	Shipping Dock 1
3e	3	Shipping Dock 2
2h	2	Random Area 1

These are your sampling sites. Please remember to **ALWAYS** assign a site to zone "1", "2", or "3".

Once you have

**Edits  
Complete**

# Current Plan

Cycle Start: 4/20/2013

Date: 5/9/2013

Sampling Plan:

2C

medium

Risk:

high

Current State:

0

Enter new data

View Reports

Zone	Site	Taken:	Goal:	Remaining:	Completed:	
1	1a	6	9	3	<input type="checkbox"/>	
	1b	6	9	3	<input type="checkbox"/>	
	1c	6	9	3	<input type="checkbox"/>	
	1d	6	9	3	<input type="checkbox"/>	
	1e	7	9	2	<input type="checkbox"/>	
	1f	3	9	6	<input type="checkbox"/>	
	1g	3	9	6	<input type="checkbox"/>	
	1h	3	9	6	<input type="checkbox"/>	
	1i	3	9	6	<input type="checkbox"/>	
	1j	3	9	6	<input type="checkbox"/>	
	2	2a	6	7	1	<input type="checkbox"/>
		2b	6	7	1	<input type="checkbox"/>
2c		3	7	4	<input type="checkbox"/>	
2d		3	7	4	<input type="checkbox"/>	
2e		3	7	4	<input type="checkbox"/>	
2f		3	7	4	<input type="checkbox"/>	
2g		2	7	5	<input type="checkbox"/>	
2h		1	7	6	<input type="checkbox"/>	
3	3a	6	8	2	<input type="checkbox"/>	
	3b	3	8	5	<input type="checkbox"/>	
	3c	3	8	5	<input type="checkbox"/>	
	3d	3	8	5	<input type="checkbox"/>	
	3e	3	8	5	<input type="checkbox"/>	
Grand Total		91	186	95	<input type="checkbox"/>	

DateTaken	DateAnaly	Zone	Site	EB_Test	S_Test	EB_pass	Current
3/1/2013	3/2/2013	1	1a	0	0	Pass	0
3/2/2013	3/2/2013	1	1a	0	0	Pass	0
3/3/2013	3/4/2013	1	1b	20	0	Fail	0
3/3/2013	3/4/2013	1	1b	0	0	Pass	0
3/3/2013	3/4/2013	1	1c	12	0	Fail	0
3/3/2013	3/4/2013	1	1c	0	0	Pass	0
3/3/2013	3/4/2013	1	1d	4	0	Pass	0
3/3/2013	3/4/2013	1	1d	12	0	Fail	0
3/4/2013	3/4/2013	1	1e	33	0	Fail	0
3/4/2013	3/4/2013	1	1e	100	0	Fail	0
3/4/2013	3/4/2013	1	1f	0	0	Pass	0
3/4/2013	3/4/2013	1	1g	0	0	Pass	0
3/4/2013	3/4/2013	1	1h	0	0	Pass	0
3/4/2013	3/4/2013	1	1i	120	0	Fail	0
3/4/2013	3/4/2013	1	1j	0	0	Pass	0
3/5/2013	3/6/2013	2	2a	0	0	Pass	0
3/5/2013	3/6/2013	2	2a	0	0	Pass	0
3/5/2013	3/6/2013	2	2b	30	0	Pass	0
3/5/2013	3/6/2013	2	2b	0	0	Pass	0
3/5/2013	3/6/2013	2	2c	90	0	Pass	0
3/6/2013	3/6/2013	2	2d	0	0	Pass	0
3/6/2013	3/6/2013	2	2e	0	0	Pass	0
3/6/2013	3/6/2013	2	2f	0	0	Pass	0
3/7/2013	3/7/2013	2	2g	190	0	Fail	0
3/7/2013	3/7/2013	3	3a	0	0	Pass	0
3/7/2013	3/7/2013	3	3a	0	0	Pass	0
3/7/2013	3/7/2013	3	3b	0	0	Pass	0
3/7/2013	3/7/2013	3	3c	355	0	Pass	0
3/7/2013	3/7/2013	3	3d	0	0	Pass	0
3/7/2013	3/7/2013	3	3e	280	0	Pass	0
3/9/2013	3/9/2013	1	1a	0	0	Pass	0
3/9/2013	3/9/2013	1	1a	0	0	Pass	0
3/9/2013	3/9/2013	1	1b	20	0	Fail	0
3/9/2013	3/9/2013	1	1b	0	0	Pass	0
3/9/2013	3/9/2013	1	1c	44	0	Fail	0
3/9/2013	3/9/2013	1	1c	0	0	Pass	0
3/9/2013	3/9/2013	1	1d	4	0	Pass	0
3/11/2013	3/11/2013	1	1d	12	0	Fail	0
3/11/2013	3/11/2013	1	1e	0	0	Pass	0
3/11/2013	3/11/2013	1	1e	66	0	Fail	0
3/11/2013	3/11/2013	1	1f	0	0	Pass	0
3/11/2013	3/11/2013	1	1g	54	0	Fail	0
3/11/2013	3/11/2013	1	1h	0	0	Pass	0
3/11/2013	3/11/2013	1	1i	8	0	Pass	0
3/11/2013	3/11/2013	1	1j	0	0	Pass	0
3/11/2013	3/11/2013	2	2a	34	0	Pass	0

3/11/2013	3/11/2013	2	2a	0	0	Pass	0
3/11/2013	3/11/2013	2	2b	30	0	Pass	0
3/13/2013	3/13/2013	2	2b	0	0	Pass	0
3/13/2013	3/13/2013	2	2c	66	0	Pass	0
3/13/2013	3/13/2013	2	2d	0	0	Pass	0
3/13/2013	3/13/2013	2	2e	0	0	Pass	0
3/13/2013	3/13/2013	2	2f	0	0	Pass	0
3/13/2013	3/13/2013	2	2h	50	0	Pass	0
3/13/2013	3/13/2013	3	3a	0	0	Pass	0
3/13/2013	3/13/2013	3	3a	0	0	Pass	0
3/13/2013	3/13/2013	3	3b	777	0	Fail	0
3/13/2013	3/13/2013	3	3c	90	0	Pass	0
3/13/2013	3/13/2013	3	3d	300	0	Pass	0
3/13/2013	3/13/2013	3	3e	28	0	Pass	0
3/16/2013	3/19/2013	1	1a	0	0	Pass	0
3/16/2013	3/19/2013	1	1a	0	0	Pass	0
3/16/2013	3/19/2013	1	1b	20	0	Fail	0
3/16/2013	3/19/2013	1	1b	0	0	Pass	0
3/16/2013	3/19/2013	1	1c	0	0	Pass	0
3/16/2013	3/19/2013	1	1c	0	0	Pass	0
3/16/2013	3/19/2013	1	1d	100	0	Fail	0
3/16/2013	3/19/2013	1	1d	12	0	Fail	0
3/16/2013	3/19/2013	1	1e	0	0	Pass	0
3/19/2013	3/19/2013	1	1e	0	0	Pass	0
3/19/2013	3/19/2013	1	1f	0	0	Pass	0
3/19/2013	3/19/2013	1	1g	0	0	Pass	0
3/19/2013	3/19/2013	1	1h	300	0	Fail	0
3/19/2013	3/19/2013	1	1i	8	0	Pass	0
3/19/2013	3/19/2013	1	1j	0	0	Pass	0
3/19/2013	3/19/2013	2	2a	0	0	Pass	0
3/19/2013	3/19/2013	2	2a	39	0	Pass	0
3/19/2013	3/19/2013	2	2b	30	0	Pass	0
3/21/2013	3/21/2013	2	2b	0	0	Pass	0
3/21/2013	3/21/2013	2	2c	444	0	Fail	0
3/21/2013	3/21/2013	2	2d	0	0	Pass	0
3/21/2013	3/21/2013	2	2e	0	0	Pass	0
3/21/2013	3/21/2013	2	2f	0	0	Pass	0
3/21/2013	3/21/2013	2	2g	654	0	Fail	0
3/21/2013	3/21/2013	3	3a	0	0	Pass	0
3/21/2013	3/21/2013	3	3a	0	0	Pass	0
3/21/2013	3/21/2013	3	3b	0	0	Pass	0
3/21/2013	3/21/2013	3	3c	452	0	Pass	0
3/21/2013	3/21/2013	3	3d	660	0	Fail	0
3/21/2013	3/21/2013	3	3e	28	0	Pass	0
3/23/2013	3/23/2013	1	1a	0	0	Pass	0
3/23/2013	3/23/2013	1	1a	0	0	Pass	0
3/23/2013	3/23/2013	1	1b	233	0	Fail	0

3/23/2013	3/23/2013	1	1b	0	0	Pass	0
3/23/2013	3/23/2013	1	1c	32	0	Fail	0
3/23/2013	3/23/2013	1	1c	0	0	Pass	0
3/23/2013	3/23/2013	1	1d	4	0	Pass	0
3/23/2013	3/23/2013	1	1d	12	0	Fail	0
3/23/2013	3/23/2013	1	1e	0	0	Pass	0
3/26/2013	3/28/2013	1	1e	0	0	Pass	0
3/26/2013	3/28/2013	1	1f	0	0	Pass	0
3/26/2013	3/28/2013	1	1g	0	0	Pass	0
3/26/2013	3/28/2013	1	1h	0	0	Pass	0
3/26/2013	3/28/2013	1	1i	8	0	Pass	0
3/26/2013	3/28/2013	1	1j	753	0	Fail	0
3/26/2013	3/28/2013	2	2a	0	0	Pass	0
3/26/2013	3/28/2013	2	2a	0	0	Pass	0
3/26/2013	3/28/2013	2	2b	30	0	Pass	0
3/26/2013	3/28/2013	2	2b	0	0	Pass	0
3/26/2013	3/28/2013	2	2c	66	0	Pass	0
3/28/2013	3/28/2013	2	2d	0	0	Pass	0
3/28/2013	3/28/2013	2	2e	529	0	Fail	0
3/28/2013	3/28/2013	2	2f	0	0	Pass	0
3/28/2013	3/28/2013	2	2h	240	0	Fail	0
3/28/2013	3/28/2013	3	3a	0	0	Pass	0
3/28/2013	3/28/2013	3	3a	0	0	Pass	0
3/28/2013	3/28/2013	3	3b	483	0	Pass	0
3/28/2013	3/28/2013	3	3c	90	0	Pass	0
3/28/2013	3/28/2013	3	3d	0	0	Pass	0
3/28/2013	3/28/2013	3	3e	28	0	Pass	0
4/1/2013	4/3/2013	1	1a	369	0	Fail	0
4/1/2013	4/3/2013	1	1a	0	0	Pass	0
4/1/2013	4/3/2013	1	1b	0	0	Pass	0
4/1/2013	4/3/2013	1	1b	0	0	Pass	0
4/1/2013	4/3/2013	1	1c	0	0	Pass	0
4/1/2013	4/3/2013	1	1c	351	0	Fail	0
4/1/2013	4/3/2013	1	1d	4	0	Pass	0
4/1/2013	4/3/2013	1	1d	0	0	Pass	0
4/1/2013	4/3/2013	1	1e	0	0	Pass	0
4/1/2013	4/3/2013	1	1e	0	0	Pass	0
4/1/2013	4/3/2013	1	1f	0	0	Pass	0
4/1/2013	4/3/2013	1	1g	0	0	Pass	0
4/6/2013	4/7/2013	1	1h	0	0	Pass	0
4/6/2013	4/7/2013	1	1i	8	0	Pass	0
4/6/2013	4/7/2013	1	1j	0	0	Pass	0
4/6/2013	4/7/2013	2	2a	0	0	Pass	0
4/6/2013	4/7/2013	2	2a	0	0	Pass	0
4/6/2013	4/7/2013	2	2b	300	0	Fail	0
4/6/2013	4/7/2013	2	2b	0	0	Pass	0
4/6/2013	4/7/2013	2	2c	50	0	Pass	0

4/6/2013	4/7/2013	2	2d	0	0	Pass	0
4/6/2013	4/7/2013	2	2e	0	0	Pass	0
4/6/2013	4/7/2013	2	2f	0	0	Pass	0
4/6/2013	4/7/2013	2	2g	10	0	Pass	0
4/6/2013	4/7/2013	3	3a	0	0	Pass	0
4/6/2013	4/7/2013	3	3a	0	0	Pass	0
4/6/2013	4/7/2013	3	3b	0	0	Pass	0
4/6/2013	4/7/2013	3	3c	90	0	Pass	0
4/6/2013	4/7/2013	3	3d	0	0	Pass	0
4/6/2013	4/7/2013	3	3e	80	0	Pass	0
4/10/2013	4/12/2013	1	1a	0	0	Pass	0
4/10/2013	4/12/2013	1	1a	0	0	Pass	0
4/10/2013	4/12/2013	1	1b	0	0	Pass	0
4/10/2013	4/12/2013	1	1b	0	0	Pass	0
4/10/2013	4/12/2013	1	1c	0	0	Pass	0
4/10/2013	4/12/2013	1	1c	0	0	Pass	0
4/10/2013	4/12/2013	1	1d	4	0	Pass	0
4/10/2013	4/12/2013	1	1d	0	0	Pass	0
4/10/2013	4/12/2013	1	1e	0	0	Pass	0
4/10/2013	4/12/2013	1	1e	0	0	Pass	0
4/10/2013	4/12/2013	1	1f	0	0	Pass	0
4/10/2013	4/12/2013	1	1g	0	0	Pass	0
4/10/2013	4/12/2013	1	1h	0	0	Pass	0
4/10/2013	4/12/2013	1	1i	8	0	Pass	0
4/12/2013	4/12/2013	1	1j	0	1	Pass	0
4/12/2013	4/12/2013	2	2a	0	0	Pass	0
4/12/2013	4/12/2013	2	2a	0	0	Pass	0
4/12/2013	4/12/2013	2	2b	30	0	Pass	0
4/12/2013	4/12/2013	2	2b	0	0	Pass	0
4/12/2013	4/12/2013	2	2c	66	0	Pass	0
4/12/2013	4/12/2013	2	2d	0	0	Pass	0
4/12/2013	4/12/2013	2	2e	0	0	Pass	0
4/12/2013	4/12/2013	2	2f	0	0	Pass	0
4/12/2013	4/12/2013	2	2h	50	0	Pass	0
4/12/2013	4/12/2013	3	3a	0	0	Pass	0
4/12/2013	4/12/2013	3	3a	0	0	Pass	0
4/15/2013	4/15/2013	3	3b	0	1	Pass	0
4/15/2013	4/15/2013	3	3c	90	0	Pass	0
4/15/2013	4/15/2013	3	3d	300	0	Pass	0
4/15/2013	4/15/2013	3	3e	28	0	Pass	0
4/20/2013	4/20/2013	1	1a	0	0	Pass	1
4/20/2013	4/20/2013	1	1a	0	0	Pass	1
4/20/2013	4/20/2013	1	1b	20	0	Fail	1
4/20/2013	4/20/2013	1	1b	0	0	Pass	1
4/20/2013	4/20/2013	1	1c	0	0	Pass	1
4/20/2013	4/20/2013	1	1c	0	0	Pass	1
4/20/2013	4/20/2013	1	1d	4	0	Pass	1



4/20/2013	4/20/2013	1	1d	12	0	Fail	1
4/20/2013	4/20/2013	1	1e	0	0	Pass	1
4/20/2013	4/20/2013	1	1e	0	0	Pass	1
4/20/2013	4/20/2013	1	1f	0	0	Pass	1
4/20/2013	4/20/2013	1	1g	0	0	Pass	1
4/20/2013	4/20/2013	1	1h	0	0	Pass	1
4/20/2013	4/20/2013	1	1i	8	0	Pass	1
4/20/2013	4/20/2013	1	1j	0	0	Pass	1
4/20/2013	4/20/2013	2	2a	0	0	Pass	1
4/24/2013	4/24/2013	2	2a	0	0	Pass	1
4/24/2013	4/24/2013	2	2b	30	0	Pass	1
4/24/2013	4/24/2013	2	2b	0	0	Pass	1
4/24/2013	4/24/2013	2	2c	66	0	Pass	1
4/24/2013	4/24/2013	2	2d	0	0	Pass	1
4/24/2013	4/24/2013	2	2e	0	0	Pass	1
4/24/2013	4/24/2013	2	2f	0	0	Pass	1
4/24/2013	4/24/2013	2	2g	80	0	Pass	1
4/24/2013	4/24/2013	3	3a	0	0	Pass	1
4/24/2013	4/24/2013	3	3a	0	0	Pass	1
4/24/2013	4/24/2013	3	3b	0	0	Pass	1
4/24/2013	4/24/2013	3	3c	90	0	Pass	1
4/24/2013	4/24/2013	3	3d	410	0	Pass	1
4/24/2013	4/24/2013	3	3e	28	0	Pass	1
4/28/2013	4/29/2013	1	1a	0	0	Pass	1
4/28/2013	4/29/2013	1	1a	0	0	Pass	1
4/28/2013	4/29/2013	1	1b	20	0	Fail	1
4/28/2013	4/29/2013	1	1b	0	0	Pass	1
4/28/2013	4/29/2013	1	1c	0	0	Pass	1
4/28/2013	4/29/2013	1	1c	0	0	Pass	1
4/28/2013	4/29/2013	1	1d	4	0	Pass	1
4/28/2013	4/29/2013	1	1d	12	0	Fail	1
4/28/2013	4/29/2013	1	1e	0	0	Pass	1
4/28/2013	4/29/2013	1	1e	0	0	Pass	1
4/28/2013	4/29/2013	1	1f	0	0	Pass	1
4/28/2013	4/29/2013	1	1g	0	0	Pass	1
4/28/2013	4/29/2013	1	1h	0	0	Pass	1
4/28/2013	4/29/2013	1	1i	8	0	Pass	1
4/28/2013	4/29/2013	1	1j	0	0	Pass	1
4/28/2013	4/29/2013	2	2a	0	0	Pass	1
4/28/2013	4/29/2013	2	2a	0	0	Pass	1
4/28/2013	4/29/2013	2	2b	30	0	Pass	1
4/28/2013	4/29/2013	2	2b	0	0	Pass	1
4/28/2013	4/29/2013	2	2c	66	0	Pass	1
4/28/2013	4/29/2013	2	2d	0	0	Pass	1
4/28/2013	4/29/2013	2	2e	0	0	Pass	1
4/28/2013	4/29/2013	2	2f	0	0	Pass	1
4/28/2013	4/29/2013	2	2h	240	0	Fail	1

4/28/2013	4/29/2013	3	3a	0	0	Pass	1
4/28/2013	4/29/2013	3	3a	0	0	Pass	1
4/28/2013	4/29/2013	3	3b	0	0	Pass	1
4/28/2013	4/29/2013	3	3c	90	0	Pass	1
4/28/2013	4/29/2013	3	3d	770	0	Fail	1
4/28/2013	4/29/2013	3	3e	28	0	Pass	1
5/2/2013	5/3/2013	1	1a	369	0	Fail	1
5/2/2013	5/3/2013	1	1a	0	0	Pass	1
5/2/2013	5/3/2013	1	1b	0	0	Pass	1
5/2/2013	5/3/2013	1	1b	0	0	Pass	1
5/2/2013	5/3/2013	1	1c	0	0	Pass	1
5/2/2013	5/3/2013	1	1c	351	0	Fail	1
5/2/2013	5/3/2013	1	1d	4	0	Pass	1
5/2/2013	5/3/2013	1	1d	0	0	Pass	1
5/2/2013	5/3/2013	1	1e	0	0	Pass	1
5/2/2013	5/3/2013	1	1e	0	0	Pass	1
5/2/2013	5/3/2013	1	1f	0	0	Pass	1
5/2/2013	5/3/2013	1	1g	0	0	Pass	1
5/6/2013	5/7/2013	1	1h	0	0	Pass	1
5/6/2013	5/7/2013	1	1i	8	0	Pass	1
5/6/2013	5/7/2013	1	1j	0	0	Pass	1
5/6/2013	5/7/2013	2	2a	0	0	Pass	1
5/6/2013	5/7/2013	2	2a	0	0	Pass	1
5/6/2013	5/7/2013	2	2b	300	0	Fail	1
5/6/2013	5/7/2013	2	2b	0	0	Pass	1
5/6/2013	5/7/2013	2	2c	50	0	Pass	1
5/6/2013	5/7/2013	2	2d	0	0	Pass	1
5/6/2013	5/7/2013	2	2e	0	0	Pass	1
5/6/2013	5/7/2013	2	2f	0	0	Pass	1
5/6/2013	5/7/2013	2	2g	10	0	Pass	1
5/6/2013	5/7/2013	3	3a	0	0	Pass	1
5/6/2013	5/7/2013	3	3a	0	0	Pass	1
5/6/2013	5/7/2013	3	3b	0	0	Pass	1
5/6/2013	5/7/2013	3	3c	90	0	Pass	1
5/6/2013	5/7/2013	3	3d	0	0	Pass	1
5/6/2013	5/7/2013	3	3e	80	0	Pass	1
5/10/2013	5/12/2013	1	1a	0	0	Pass	0
5/10/2013	5/12/2013	1	1a	0	0	Pass	0
5/10/2013	5/12/2013	1	1b	0	0	Pass	0
5/10/2013	5/12/2013	1	1b	0	0	Pass	0
5/10/2013	5/12/2013	1	1c	0	0	Pass	0
5/10/2013	5/12/2013	1	1c	0	0	Pass	0
5/10/2013	5/12/2013	1	1d	4	0	Pass	0
5/10/2013	5/12/2013	1	1d	0	0	Pass	0
5/10/2013	5/12/2013	1	1e	0	0	Pass	0
5/10/2013	5/12/2013	1	1e	0	0	Pass	0
5/10/2013	5/12/2013	1	1f	0	0	Pass	0

5/10/2013	5/12/2013	1	1g	0	0	Pass	0
5/10/2013	5/12/2013	1	1h	0	0	Pass	0
5/10/2013	5/12/2013	1	1i	8	0	Pass	0
5/10/2013	5/12/2013	1	1j	0	0	Pass	0
5/10/2013	5/12/2013	2	2a	0	0	Pass	0
5/10/2013	5/12/2013	2	2a	0	0	Pass	0
5/12/2013	5/12/2013	2	2b	30	0	Pass	0
5/12/2013	5/12/2013	2	2b	0	0	Pass	0
5/12/2013	5/12/2013	2	2c	66	0	Pass	0
5/12/2013	5/12/2013	2	2d	0	0	Pass	0
5/12/2013	5/12/2013	2	2e	0	0	Pass	0
5/12/2013	5/12/2013	2	2f	0	0	Pass	0
5/12/2013	5/12/2013	2	2h	50	0	Pass	0
5/12/2013	5/12/2013	3	3a	0	0	Pass	0
5/12/2013	5/12/2013	3	3a	0	0	Pass	0
5/15/2013	5/15/2013	3	3b	0	0	Pass	0
5/15/2013	5/15/2013	3	3c	90	0	Pass	0
5/15/2013	5/15/2013	3	3d	300	0	Pass	0
5/15/2013	5/15/2013	3	3e	28	0	Pass	0
5/20/2013	5/20/2013	1	1a	0	0	Pass	0
5/20/2013	5/20/2013	1	1a	0	0	Pass	0
5/20/2013	5/20/2013	1	1b	20	0	Fail	0
5/20/2013	5/20/2013	1	1b	0	0	Pass	0
5/20/2013	5/20/2013	1	1c	0	0	Pass	0
5/20/2013	5/20/2013	1	1c	0	0	Pass	0
5/20/2013	5/20/2013	1	1d	4	0	Pass	0
5/20/2013	5/20/2013	1	1d	12	0	Fail	0
5/20/2013	5/20/2013	1	1e	0	0	Pass	0
5/20/2013	5/20/2013	1	1e	0	0	Pass	0
5/20/2013	5/20/2013	1	1f	0	0	Pass	0
5/20/2013	5/20/2013	1	1g	0	0	Pass	0
5/20/2013	5/20/2013	1	1h	0	0	Pass	0
5/20/2013	5/20/2013	1	1i	8	0	Pass	0
5/20/2013	5/20/2013	1	1j	0	0	Pass	0
5/20/2013	5/20/2013	2	2a	0	0	Pass	0
5/20/2013	5/20/2013	2	2a	0	0	Pass	0
5/24/2013	5/24/2013	2	2b	30	0	Pass	0
5/24/2013	5/24/2013	2	2b	0	0	Pass	0
5/24/2013	5/24/2013	2	2c	66	0	Pass	0
5/24/2013	5/24/2013	2	2d	0	0	Pass	0
5/24/2013	5/24/2013	2	2e	0	0	Pass	0
5/24/2013	5/24/2013	2	2f	0	0	Pass	0
5/24/2013	5/24/2013	2	2g	80	0	Pass	0
5/24/2013	5/24/2013	3	3a	0	0	Pass	0
5/24/2013	5/24/2013	3	3a	0	0	Pass	0
5/24/2013	5/24/2013	3	3b	0	0	Pass	0
5/24/2013	5/24/2013	3	3c	90	0	Pass	0

5/24/2013	5/24/2013	3	3d	410	0	Pass	0
5/24/2013	5/24/2013	3	3e	28	0	Pass	0
5/24/2013	5/24/2013	1	1a	0	0	Pass	0
5/24/2013	5/24/2013	1	1a	0	0	Pass	0
5/24/2013	5/24/2013	1	1b	20	0	Fail	0
5/24/2013	5/24/2013	1	1b	0	0	Pass	0
5/24/2013	5/24/2013	1	1c	0	0	Pass	0
5/24/2013	5/24/2013	1	1c	0	0	Pass	0
5/24/2013	5/24/2013	1	1d	4	0	Pass	0
5/24/2013	5/24/2013	1	1d	12	0	Fail	0
5/24/2013	5/24/2013	1	1e	0	0	Pass	0
5/24/2013	5/24/2013	1	1e	0	0	Pass	0
5/24/2013	5/24/2013	1	1f	0	0	Pass	0
5/24/2013	5/24/2013	1	1g	0	0	Pass	0
5/24/2013	5/24/2013	1	1h	0	0	Pass	0
5/24/2013	5/24/2013	1	1i	8	0	Pass	0
5/28/2013	5/29/2013	1	1j	0	0	Pass	0
5/28/2013	5/29/2013	2	2a	0	1	Pass	0
5/28/2013	5/29/2013	2	2a	0	0	Pass	0
5/28/2013	5/29/2013	2	2b	30	0	Pass	0
5/28/2013	5/29/2013	2	2b	0	0	Pass	0
5/28/2013	5/29/2013	2	2c	66	0	Pass	0
5/28/2013	5/29/2013	2	2d	0	0	Pass	0
5/28/2013	5/29/2013	2	2e	0	0	Pass	0
5/28/2013	5/29/2013	2	2f	0	0	Pass	0
5/28/2013	5/29/2013	2	2h	240	0	Fail	0
5/28/2013	5/29/2013	3	3a	0	0	Pass	0
5/28/2013	5/29/2013	3	3a	0	0	Pass	0
5/28/2013	5/29/2013	3	3b	0	0	Pass	0
5/28/2013	5/29/2013	3	3c	90	0	Pass	0
5/28/2013	5/29/2013	3	3d	770	0	Fail	0
5/28/2013	5/29/2013	3	3e	28	0	Pass	0
4/22/2013	4/23/2013	1	1e	0	0	Pass	1

# Current Cycle

## Current 1 EB-Test

Total #		EB_pass		Total
Zone	Site	Fail	Pass	Samples
1	1a	2	2	2
	1b		1	2
	1c		2	2
	1d		1	2
	1e		3	3
	1f		1	1
	1g		1	1
	1h		1	1
	1i		1	1
	1j		1	1
	<b>1 Total</b>		<b>2</b>	<b>14</b>
2	2a	2	2	2
	2b		2	2
	2c		1	1
	2d		1	1
	2e		1	1
	2f		1	1
	2g		1	1
<b>2 Total</b>		<b>9</b>	<b>9</b>	
3	3a	2	2	2
	3b		1	1
	3c		1	1
	3d		1	1
	3e		1	1
<b>3 Total</b>		<b>6</b>	<b>6</b>	
<b>Total Samples</b>		<b>2</b>	<b>29</b>	<b>31</b>

## Current 1 S-Test

Total #		S_Test		Total
Zone	Site	Pass	Total	Samples
1	1a	2	2	2
	1b		2	2
	1c		2	2
	1d		2	2
	1e		3	3
	1f		1	1
	1g		1	1
	1h		1	1
	1i		1	1
	1j		1	1
	<b>1 Total</b>		<b>16</b>	<b>16</b>
2	2a	2	2	2
	2b		2	2
	2c		1	1
	2d		1	1
	2e		1	1
	2f		1	1
	2g		1	1
<b>2 Total</b>		<b>9</b>	<b>9</b>	
3	3a	2	2	2
	3b		1	1
	3c		1	1
	3d		1	1
	3e		1	1
<b>3 Total</b>		<b>6</b>	<b>6</b>	
<b>Total Samples</b>		<b>31</b>	<b>31</b>	

Main Menu

Curren

**Total "S" sample fail limit per month**

if # of failed samples > 1 sample per zone (1, 2 or both 1.2), then shift to higher risk sampling plan (low --> High or med. --> high) for 1 months, Till you receive a negative result for 2 week.

Initial Sample Plan	Plant Volume	Product/ process risk	Salmonella samples per month				Total	State 1		
			Total	zone 1	zone 2	zone 3		z1	z2	z3
1A	low	low	30	15	9	6	1	52	40	27
1B	low	medium	60	30	18	12	1	52	40	27
1C	low	high	90	45	27	18	1	52	40	27
2A	medium	low	60	30	18	12	1	90	54	36
2B	medium	medium	120	60	36	24	1	90	54	36
2C	medium	high	180	90	54	36	1	90	54	36
3A	high	low	90	45	27	18	1	135	81	54
3B	high	medium	180	90	54	36	1	135	81	54
3C	high	high	270	135	81	54	1	135	81	54
			refer to PivotTable summary:					Mo-Salm		

EB samples per month				Total EB sample fail limit per month from all zones	State 2			State 3			State 4		
Total	zone 1 (50%)	zone 2 (30%)	zone 3 (20%)		Total	z1	z2	z3	z1	z2	z3	z1	z2
30	15	9	6	10	30	18	12				14	8	5
60	30	18	12	20	60	36	24	30	18	12	27	16	11
90	45	27	18	30	90	54	36	45	27	18	41	24	16
60	30	18	12	30	60	36	24				27	16	11
120	60	36	24	50	120	72	48	60	36	24	54	32	22
180	90	54	36	70	180	108	72	90	54	36	81	49	32
90	45	27	18	40	90	54	36				41	24	16
180	90	54	36	70	180	108	72	90	54	36	81	49	32
270	135	81	54	120	270	162	108	135	81	54	122	73	49
refer to PivotTable summary:					Mo-EB			Mo-EB			Mo-EB		

Total EB sample fail limit per month from all zones

If # failed samples in a month > than limit, then increase EB samples by 2X each zone for 1 month till you receive under norm results

if # of failed samples > limit for 3 consecutive months, then shift to higher risk sampling plan (low --> med., or med. --> high) for 1 months till you receive under norm results

if # of failed samples < limit for 3 consecutive months, then you may reduce total # of sample analyses by 10% (by pooling samples) if you have a good history of your results

State 2

State 3

State 4

Total	zone 1 (50%)	zone 2 (30%)	zone 3 (20%)	Total	z1	z2	z3	z1	z2	z3	z1	z2	z3
30	15	9	6	10	30	18	12				14	8	5
60	30	18	12	20	60	36	24	30	18	12	27	16	11
90	45	27	18	30	90	54	36	45	27	18	41	24	16
60	30	18	12	30	60	36	24				27	16	11
120	60	36	24	50	120	72	48	60	36	24	54	32	22
180	90	54	36	70	180	108	72	90	54	36	81	49	32
90	45	27	18	40	90	54	36				41	24	16
180	90	54	36	70	180	108	72	90	54	36	81	49	32
270	135	81	54	120	270	162	108	135	81	54	122	73	49

refer to PivotTable summary: Mo-EB Mo-EB Mo-EB