Food and Water
Collection, Packaging, and Laboratory Testing Guidelines

Training 2006
Updated April 2007
LEARNING OBJECTIVES:

• Effectively Communicate the Response of Food and Water Testing to Appropriate Public Health Professionals
• Describe and Properly Use the Contents of a Food & Water Collection Kit
• Collect Food and Water Samples Using Aseptic & Appropriate Techniques
• Provide Appropriate Documentation for Food and Water Samples
• Properly Package & Transport Food & Water Samples to the State’s Division of Laboratory Services (DLS)
• Display General Knowledge of Food & Water Testing Methodology
OUTLINE of TRAINING

• FOOD COLLECTION PROCESS:
  – GENERAL INFORMATION
  – COLLECTION KIT
  – FOOD KIT USAGE
  – PACKAGING & SHIPPING
  – LABORATORY TESTING

• WATER COLLECTION PROCESS:
  – GENERAL INFORMATION
  – COLLECTION KIT
  – WATER KIT USAGE
  – PACKAGING & SHIPPING
  – LABORATORY TESTING

• SPECIAL CONSIDERATIONS:
  – CHEMICAL
  – FORENSIC TESTING
  – BIOLOGICAL AGENT
THE NEED FOR TRAINING & INFORMATION:
FREQUENT PROBLEMS

- COMMUNICATION
- SUBMISSION
  - NO ADVANCE NOTICE (Food)
  - APPROVAL?? (Food)
- VAGUE TEST REQUEST
- QUANTITY
  - INADEQUATE
  - INCORRECT
- INFREQUENT SAMPLING
  - where is my food kit
  - recall how to properly collect, package, & ship
KPHL – FOOD MICROBIOLOGY

KY Public Health Laboratory

FOOD COLLECTION, PACKAGING, & LABORATORY TESTING GUIDELINES
FOOD COLLECTION – WHY?

- ROUTINE SURVEILLANCE
  (manufacturing, retail, facility)

- CONSUMER COMPLAINT:
  ILLNESS or INJURY

- VISIBLE CONTAMINATION:
  - Object (foreign or known)
  - Unusual substance
  - Unusual appearance
  - Animal droppings, tracks, signs, etc.

- UNUSUAL TASTE

- ODOR
FOOD COLLECTION KIT

- LOCATE YOUR KIT

- CHECK THE CONTENTS PERIODICALLY

- REPLACE EXPIRED ITEMS

- ORDER KIT & ITEMS
  (some items may be easily replaced at the LHD such as alcohol pads, sterile tongue depressors, sterile specimen cups, etc.)
FOOD COLLECTION KIT

Order kits on-line:
http://chfs.ky.gov/dph/info/lab....... (Kit contains Collection items, Information, & Forms)

Kits may also be ordered by calling the lab at 502-564-4446.
FOOD COLLECTION KIT CONTENTS

Food Collection Kit Basics:

- FORMS
- INFORMATION

- COLLECTION SUPPLIES
  and ACCESSORIES

- COLLECTION CONTAINERS
THIS FOOD COLLECTION KIT CONTENT SHEET:

THIS FOOD SAMPLE COLLECTION KIT CONTAINS THE FOLLOWING:

I. FORMS:
LAB
(1) “SUGGESTED PROTOCOL FOR USE OF FOOD COLLECTION KIT” LAB 302h (REV 3-2006)
(4) “SAMPLE COLLECTION DATA AND ANALYSIS REPORT” LAB 304 (REV 3-00)

FOOD SAFETY
(2) “RETAIL FOOD ESTABLISHMENT REPORT” DFS 208 (REV 05/05)
(2) “NOTICE AND ORDER OF QUARANTINE / VOLUNTARY DESTRUCTION” DFS 222
(2) “QUARANTINED” TAG DFS 223
(1) “RECORD OF COMPLAINT AND INVESTIGATION” DFS 216 (food manufacturing)

EPIDEMIOLOGY (contact epidemiologists for suspected outbreak & investigation)
Electronic Foodborne Outbreak Reporting System (eFORS) - web-based Form 52-13
Epidemiologist Investigation - http://www.cdc.gov/foodborneoutbreaks/toolkit.htm

II. COLLECTION CONTAINERS:

“WHIRL-PAK” STERILE PLASTIC BAGS (5)
“SAMPLE BAG” - POLYETHYLENE LINED (1)
4 oz - STERILE SCREW CAPPED SPECIMEN CUP
8 oz - HINGED SAMPLING VIAL - POLYPROPYLENE
4 oz - GLASS BOTTLE (LIQUIDS for CHEMICAL ANALYSIS) - fill to top / no air bubbles
ALUMINUM FOIL SHEET (use to wrap FISH for CHEMICAL ANALYSIS)

III. SAMPLE COLLECTION SUPPLIES: ACCESSORIES:
STERILE PLASTIC TABLESPOON
STERILE PLASTIC SCOOP
STERILE TONGUE DEPRESSORS (6)
STERILE SAMPLING KNIFE
ALCOHOL PADS (10)
MARKER
PAPER BAGS - NON-STERILE

“OFFICIAL” SAMPLE SEALING TAPE (REQUIRES - SAMPLE#, DATE, SIGNATURE, AGENCY) DFS 224

ITEMIZED LIST CATEGORIZED INTO 3 SECTIONS (I. II. III.)
SECTION I. FORM – LABORATORY
LAB FORM 504 (Rev. 4-4-05)
LAB FORM 504

Thoroughly Complete:

- Sample ID
- Date of Collection
- Collector
- Procured from
- Sample Description

- Requested Lab Analysis
  (Bacteriological, Chemical, Other)

- Signature of Submitter
Consult First
Food Safety Branch
before sending food for testing
(502) 564-7181

Bacteriological analysis must be determined based upon Clinical findings:

- Clinical Symptoms
- Onset of Illness
- Duration of Illness
- Patient Testing (culture)

Epidemiology: (502) 564-3261

Chemical analysis; narrow down
Suspect Chemical with thorough investigation & Interview
Bacteriological Request cannot be a “shot-gun” (test for all) approach (Do not check all of the bacteria boxes)

NOTE: KPHL is not a diagnostic lab - confirms & supports diagnostic and clinical testing) 

NOR

a physician’s script of “culture the sample and see what grows” 
(Do not check all of the bacteria boxes)

THEREFORE:

Consult your Regional Epidemiologist / Epi Nurse or State Epidemiologist (502-564-3261) to narrow down to 1 or 2 suspect bacterial pathogens

Specific food “pathogens” (cause disease or illness) must be requested based upon thorough investigation, interviewing, clinical findings, and epidemiology of illness (aids in determining the test request of 1 or 2 suspect bacteriological pathogens)
### SECTION I. FORMS - FOOD SAFETY

**BRANCH DFS – 216 (4-95 - Complaint)**

#### RECORD OF COMPLAINT AND INVESTIGATION

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**KENTUCKY UNBRIDLED SPIRIT**

Cabinet for Health and Family Services
I. FORMS - FOOD SAFETY BRANCH
DFS – 222 (5-94) – Quarantine/Destruction

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<th>Article ID</th>
<th>Quantity</th>
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**NOTICE AND ORDER OF QUARANTINE / VOLUNTARY DESTRUCTION**

**Article ID**: DFS-222 (5-94)

**Date**: 3-31-06

**Permit No.**: 51-100

**Quarantine Code**: No Quantity

**Quarantine Code**: No Quantity

**Serial Code**: No Quantity

**Pursuant to the Provisions of**: 605 KYS 100-100 Regulations Use and Control of Rodent

**In Possession**: Located at Joe's Restaurant located at 1101 2nd street, Anywhere, KY

**And is Being Quarantined**: On this 31st Day of March 2006 at 10:00 AM

**Estimated Reported Value**: $10.06

**Total Quantity**: 10

**LBS**: All persons are warned not to remove or dispose of the above-described articles by sale or otherwise until permission for removal or disposal is given by a duly authorized agent of the cabinet for human resources or by a court of competent jurisdiction.

**Acknowledgment of**:

Q1 Notice and Order of Quarantine by Owner or Person in Possession, or Agent thereof, if available.

Q2 Voluntary Destruction by the owner of the articles identified above. I (we) agree to the voluntary destruction of the listed material and hereby release the Kentucky cabinet for human resources and their members and agents from any and all responsibility.

Q3 Date and Method of Destruction:

Q4 Division: The above article(s) are released for division only for as agreed and certified by my signature below.

Signature: Date: 1101 2nd street

Inspector's Signature: City or Town

---

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Signature: Date: 1101 2nd street

Inspector's Signature: City or Town
I. FORMS - FOOD SAFETY BRANCH
DFS – 223 (Rev. 10-90) – Quarantine Tags

**QUARANTINED**

Kentucky Department for Health Services

DFS 223 (Rev. 10-90)

**QUARANTINED**

PURSUANT TO KRS-217.115
DO NOT REMOVE OR DISPOSE WITHOUT PERMISSION
BY ORDER OF THE DEPARTMENT FOR HEALTH SERVICES

AUTHORIZED AGENT

DATE

**FRONT VIEW**

**BACK VIEW**
I. FORMS - FOOD SAFETY BRANCH

DFS – 208 (REV. 05/05) - Retail
Record of Complaint & Investigation:

- Food complaints and illnesses resulting from commercial food products.
- Complete the form in detail
- FAX Form (502-564-0398) and/or call Food Safety Branch (502-564-7181) immediately to determine if samples should be collected and submitted to the State Laboratory.
- Distribute a copy of this form to the following: Food Safety Branch, LHD, and one copy for EMIS entry.
- Make a copy to the complainant only upon request
Each food sample must have a completed Official State Seal attached.

Call the Food Safety Branch (FSB) to order additional seals & FSB Forms 502-564-7181
Each Sample Must Have A Seal
Foodborne illness: transmitted to humans in which 2 or more people experience the same illness symptoms after eating a common food

FORMS: RDDR or http://www.cdc.gov/foodborneoutbreaks/toolkit.htm
THIS FOOD COLLECTION KIT CONTAINS THE FOLLOWING – FOCUSING ON SECTION II

I. FORMS:
“SUGGESTED PROTOCOL FOR USE OF FOOD COLLECTION KIT” LAB 5021 (REV 3-2006)
(4) “SAMPLE COLLECTION DATA AND ANALYSIS REPORT” LAB 564 (REV 5-00)
(2) “RETAIL FOOD ESTABLISHMENT REPORT” DFS 208 (REV 6-85)
(2) “NOTICE AND ORDER OF QUARANTINE / VOLUNTARY DESTRUCTION” DFS 222 (REV 5-94)
(2) “QUARANTINED” TAG DFS 222 (REV 10-90)
(1) “RECORD OF COMPLAINT AND INVESTIGATION” DFS 216 (4-95) (food manufacturing)
(10) “FOODBORNE DISEASE QUESTIONNAIRE” EPID-212
(2) “ATTACK TABLE” EPID

II. COLLECTION CONTAINERS:
“WHIRL-PACK” STERILE PLASTIC BAGS (5)
“SAMPLE BAG” – POLYETHYLENE LINED (5)
4 oz - STERILE SCREW CAPPED SPECIMEN CUP
8 oz - INGRED SAMPLING VIAL – POLYPROPYLENE
4 oz - GLASS BOTTLE (LIQUIDS for CHEMICAL ANALYSIS)
ALUMINUM FOIL SHEET (FISH for CHEMICAL ANALYSIS)

III. SAMPLE COLLECTION SUPPLIES/ACCESSORIES:
STERILE PLASTIC TABLESPOON
STERILE PLASTIC SCOOP
STERILE TONGUE DEPRESSEORS (6)
STERILE SAMPLING KNIFE
ALCOHOL PADS (10)
MARKER
PAPER BAGS – NON-STERILE
“OFFICIAL” SAMPLE SEALING TAPE (REQUIRES – SAMPLE#, DATE, SIGNATURE, AGENCY)
II. COLLECTION CONTAINERS:

- “WHIRL-PAK” STERILE PLASTIC BAGS
- “SAMPLE BAG” – POLYETHYLENE LINED
- 4 oz - SCREW CAP SPECIMEN CUP
- 8 oz - HINGED SAMPLING VIAL (POLYPROPYLENE)
- ALUMINUM FOIL SHEET (FISH for CHEMICAL ANALYSIS)
- 4 oz - *GLASS BOTTLE (BEVERAGE - CHEMICAL ANALYSIS)

* Bottles in the new food kits are washed with soap & water and then rinsed in acetone. The lids are TEFLOWN, not plastic (therefore, don’t require foil between lid and jar mouth). These conditions make them appropriate for chemical analysis and will not interfere with sensitive analytical instruments.
FOOD COLLECTION KIT CONTAINERS
FOCUSING ON SECTION III OF THE FOOD COLLECTION KIT:

THIS FOOD SAMPLE COLLECTION KIT CONTAINS THE FOLLOWING:
Lab 5021 (Rev 3/2006)

I. FORMS:
“SUGGESTED PROTOCOL FOR USE OF FOOD COLLECTION KIT” LAB 5021 (REV 3-2006)
(4) “SAMPLE COLLECTION DATA AND ANALYSIS REPORT” LAB 564 (REV 5-00)
(2) “RETAIL FOOD ESTABLISHMENT REPORT” DFS 208 (REV 05/05)
(2) “NOTICE AND ORDER OF QUARANTINE / VOLUNTARY DESTRUCTION” DFS 222 (REV 5-94)
(2) “QUARANTINED” TAG DFS 222 (REV 10-90)
(1) “RECORD OF COMPLAINT AND INVESTIGATION” DFS 216 (4-95) (food manufacturing)
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III. SAMPLE COLLECTION SUPPLIES/ ACCESSORIES:
STERILE PLASTIC TABLESPOON
STERILE PLASTIC SCOOP
STERILE TONGUE DEPRESSORS (6)
STERILE SAMPLING KNIFE
ALCOHOL PADS (10)
MARKER
PAPER BAGS – NON-STERILE
“OFFICIAL” SAMPLE SEALING TAPE (REQUIRES - SAMPLE, DATE, SIGNATURE, AGENCY)

ITEMIZED LIST CATEGORIZED INTO 3 SECTIONS
III. SAMPLE COLLECTION SUPPLIES:

- STERILE PLASTIC TABLESPOON
- STERILE PLASTIC SCOOP (4oz)
- STERILE TONGUE DEPRESSORS
- STERILE SAMPLING KNIFE
- ALCOHOL PADS
- PAPER BAGS NON - STERILE
- ALUMINUM FOIL — (wrap fish for chemical analysis) or (liquid drinks - place between lid & mouth if the lid is plastic - for chemical analysis – no air or bubbles )
- MARKER
- “OFFICIAL” SAMPLE SEALING TAPE
  (REQUIRES - SAMPLE#, DATE, SIGNATURE, AGENCY)
Collecting Food Samples

WASH HANDS BEFORE YOU COLLECT SAMPLES

“Aseptic” food collection is promoted by the use of gloves

Samples must be collected in aseptic manner to avoid contamination by hands or other non-sterile surfaces.
Collection of Food Samples

WHEN COLLECTING FOOD SAMPLES CONSIDER THE FOLLOWING 1ST:

• Whenever possible, Collect & send the ENTIRE leftover, unconsumed portion of the suspected contaminated food product.

• Samples should represent the entire lot of material or suspect food under evaluation.
Collection of Food Samples - Quantity

If, the entire leftover, unconsumed portion of the suspected contaminated food product cannot be collected, use the following guide:

**Liquids & Semi-solids**
Collect at least 100 grams (4 oz)

**Dry Food**
Collect 200-400 Grams

**Meat**
Collect at least 100 grams (Use sterile knife to cut meat. Sterile tongue depressors are useful for ground meat)
Collection of Food Samples - Quantity

Why is QUANTITY Important?

Because.....

!!! 25 grams is the Minimum for most analysis for just 1 food pathogen

!!! Some require 50 grams or more.

Cabinet for Health and Family Services
Collecting Food Samples Using a Whirl-Pak®

1. Tear off top of bag along perforation.
2. Use pull tabs to open bag.
3. Place sample in bag.
4A. Hold bag by wire ends and whirl three times to close.
4B. Or, fold the tab over tightly three times to close.
5. Bend wire ends onto bag.
A food sample should be taken in the center & sides of a bulk food container.

If a food product is composed of several parts or components, sample the components separately in a way as to eliminate any potential of cross-contamination.
CONTROL SAMPLES

If applicable, a “CONTROL Sample should be collected:

unopened / uncompromised package of the same food product out of the same manufacturing lot

or

“like coded” sample collected at random from available units of a product
Control Food Samples- Prepared Foods

Control food samples of “PREPARED” foods should be verified with the food establishment that the food was prepared the same way as the suspect food sample,

But not part of the outbreak or complaint
Timely Food Collection Is Important!!!

Delays in food collection coupled with “stressed” food conditions makes food pathogen survival & recovery for testing – difficult!!

STRESSED FOOD CONDITIONS:

• Bacteria in food may have been dried, frozen, chilled, heated to high temperatures, chilled again, heated again, and so on…… and so on……

• Some processed foods contain preservatives that prolong shelf life, but can inhibit bacterial growth

• Numerous other competing bacteria may be contained in the food that may grow and multiply, hindering the recovery and survival of bacterial food pathogens.
FOOD SAMPLE NOTIFICATION for LABORATORY TESTING

Why is it important to **NOTIFY** the Lab **PRIOR** to submission of food samples:

- Food pathogen testing is time consuming and involved; the lab needs time to plan & prepare
- **Each** food pathogen has a unique protocol of media (specific nutrients & pH) and incubation temperature. Media is made on demand because commercial media is expensive & has short expirations. Making media is time consuming!
Food Samples - Packaging

Maintain the original conditions under which the food was held

- Food products canned or in a dry condition, no particular precautions are necessary
- Food products under refrigeration or frozen, should be transported in such a way as to arrive in the laboratory in an unchanged condition
- Thawed samples of frozen products must be kept refrigerated; **DO NOT REFREEZE**
- Food Samples collected at a high temperature, refrigerate before packing for transporting
Protect Food Samples During TRANSPORT & SHIPPING

Proper packaging helps maintain correct temperature & pH critical to bacterial survival

Sample must be kept cold during transport

Cabinet for Health and Family Services
Food Samples - SHIPPING

Ship/Transport samples to the laboratory as rapidly as possible.

Ideally, food samples should arrive at the State Lab by the next day after collection.

If mailed, send by overnight delivery service.

But......Don’t send samples over weekends or Holidays unless advised to do so.
Valid, Reliable, and Significant Laboratory testing results and their interpretation depends upon the following:

Proper

♦ Collection

♦ Holding conditions – proper packaging

♦ Timeliness of shipping & delivery
Your Field Handbook

a good source of information on food collection issues

Field Handbook on line: www.chfs.ky.gov/dph/sanitarians.htm

Prepared by the Kentucky Registered Sanitarian Examining Committee of the Department for Public Health.
KPHL offers analysis for a number of food pathogens

Most commonly tested food pathogens:

- *Staphylococcus aureus*
- *Salmonella* species
- *Listeria* species
- *Bacillus cereus*
- *E. coli* O157:H7
- *Campylobacter*

Analysis for other organisms in food must be decided on a case by case basis. Call the KPHL Food Microbiology lab immediately for consultation at 502-564-4446
Food Pathogens - Overview

- Salmonella
- E.coli & E.coli O157:H7
- Listeria
- Campylobacter
- Staph aureus
- Bacillus cereus
Salmonella species

- It is estimated that 2 – 4 million cases of Salmonellosis occur in the U. S. annually.

- Widespread occurrence in animals, especially in poultry and swine.

- Environmental sources:
  
  water, soil, insects, factory surfaces, kitchen surfaces, animal feces, raw meats, raw poultry, and raw sea foods, to name only a few.

USFDA CFSAN Bad Bug Book

Cabinet for Health and Family Services
Salmonella species

- Foods implicated have been raw meats, poultry, eggs, milk and dairy products, fish, shrimp, frog legs, yeast, coconut, sauces and salad dressings, cake mixes, cream-filled desserts and toppings, dried gelatin, peanut butter, cocoa, and chocolate.

- Salmonella has also been isolated from both the inside and outside of eggs.

USFDA CFSAN Bad Bug Book
Salmonella species

- Confirmation of Salmonella is by biochemical and serological methods.

- The presence or absence of Salmonella species is reported.

Salmonella growing on HE agar
E. coli & E. coli O157:H7

- E. coli is a normal inhabitant of animal & human intestines; suppressing harmful bacteria and synthesizing vitamins.

- E. coli is the dominant species found in feces.

- Some E. coli strains cause human illness such as E. coli O157:H7.

- E. coli O157:H7 produces large quantities of 1 or more potent toxins that causes severe damage to the intestine lining and hemorrhagic colitis.

SUSPECT E. coli:

- Improper hand washing - food prep/process
- Sewage contamination
- Contaminated fertilizer
- Livestock in close proximity to crops/produce

USFDA CFSAN Bad Bug Book
E. coli O157:H7

“SUSPECT” E. coli O157:H7 Foods:

- Meats; Undercooked or raw hamburger (majority of nearly all documented outbreaks)
- Raw milk
- Produce contaminated with sewage
- Our State lab isolated E. coli O157:H7 from unpasteurized apple cider during routine surveillance testing of the product for the Food Safety Branch.
E. coli O157:H7 growing on Sorbitol MacConkey agar

**Presence** or **Absence** of *E. coli* O157:H7 is reported.
Listeria species

- Listeria is very widespread in the environment.

- Listeria is quite hardy and resists the damaging effects of freezing, drying and heat.

- Listeria has the ability to grow at temperatures as low as 3º C permitting multiplication in refrigerated foods. Therefore, package & ship foods suspected to contain Listeria at refrigerated temperatures
Listeria species

**SUSPECT LISTERIA:**

- milk, cheese, ice cream, uncooked vegetables, fermented raw-meat sausages, raw and cooked poultry, raw meats (all types), and raw and smoked fish.

- An episode associated with consumption of coleslaw was linked with cabbage from a farm using sewage fertilizer.

USFDA CFSAN Bad Bug Book
Listeria species

Hydrolysis of esculin causes a black halo around colonies of L. monocytogenes on Oxford and Palcam agars.

**Presence** or **Absence** of Listeria is reported.
Campylobacter species

Campylobacter is thought to be the leading bacterial cause of sporadic (non-clustered) cases of diarrheal disease in the U. S.

Outbreaks have been associated with un-treated water, raw clams, and with raw milk.
Campylobacter species

- Campylobacter jejuni is thought to be the leading cause of bacterial diarrheal illness in the U.S., but other species of Campylobacter can cause illness in humans.

- C. jejuni frequently contaminates raw chicken, inadequately cooked chicken & re-contaminated chicken. Surveys show that 20 to 100% of retail chickens are contaminated.

- Raw milk has been a source of infections.

- Note: Listeria is often carried by healthy cattle and by flies.

USFDA CFSAN Bad Bug Book
Campylobacter species

- Typical colonies of *Campylobacter jejuni* on Campy Blood Agar.

- The presence or absence of *Campylobacter* species is reported.
Staphylococcus aureus

- Staphylococci exist in air, dust, sewage, water, milk and food or on food equipment, environmental surfaces, humans and animals.

- Staphylococci are present in the nasal passages and throats and on the hair and skin of 50% or more of healthy individuals.

- Most often the source of food contamination in food poisoning outbreaks with S. aureus are food handlers or food processors.

Compendium of Methods for the Microbiological Examination of Food 3rd Edition, 1992
USFDA CFSAN Bad Bug Book
Staphylococcus aureus

Typical contaminated foods / conditions:

- Meat & meat products, milk & dairy products, egg products, salads (egg, tuna, chicken, potato, and macaroni), sandwich fillings, and bakery products such as cream-filled pastries, cream pies, and chocolate éclairs.

- Foods requiring considerable handling during preparation & are kept at slightly elevated temperatures after preparation

- Foods - un-refrigerated or improperly refrigerated for prolonged periods of time, allowing the organisms time to reproduce and to make toxin.

Compendium of Methods for the Microbiological Examination of Food 3rd Edition, 1992
USFDA CFSAN Bad Bug Book
Staphylococcus aureus

- Staphylococcal food poisoning usually does not occur unless *S. aureus* bacteria are present in the food in large numbers.

- **INFECTIVE DOSE** - a toxin dose of less than 1.0 microgram in contaminated food will produce symptoms of staphylococcal intoxication. This toxin level is reached when *S. aureus* populations exceed 100,000 per gram.

- Therefore, testing for *S. aureus* includes enumeration of colony forming units per gram of food.
Serial dilutions are made starting with a 1:10 dilution of the food. Dilutions are plated either to Baird Parker media or 3M Staph Express films. The number of S. aureus per gram is reported.
Bacillus cereus

- Bacillus cereus is widely distributed in Nature and can be isolated from a variety of foods.

- However, unless it is able to multiply, its presence is not significant to human health.

- The presence of large numbers of B. cereus (>10^6 organisms per gram) in food is indicative of active growth and proliferation of the organism consistent with a potential hazard to health.
Bacillus cereus

- B. cereus can cause 2 distinct illness syndromes; nausea & vomiting (emetic) diarrhea & abdominal pain (diarrheal)

- 95% of all emetic syndrome cases are related to fried or cooked rice.

- Meat based dishes, soups, vegetables, puddings & sauces contaminated with B. cereus has caused Diarrheal syndrome

- In almost all cases, the implicated food has been held for too long at unsatisfactory storage temperatures.
B. cereus is tested quantitatively for the number of colony forming units per gram of food.

Dilutions are made starting with a 1:10 dilution of the food. Dilutions are plated to MYEP agar. The number of B. cereus per gram is reported.
Miscellaneous Food Testing
Indicator: “Cleanliness” of Food

Coliforms, Aerobic counts, Mold & Yeast counts give an indication of the general cleanliness of food.

Honey & Sorghum production
Water Microbiology
Collection, Packaging, & Laboratory Perspective
2006
The Water lab at the KY Public Health Laboratory in Frankfort operates under EPA certification.
The KY Public Health Laboratory (KPHL) in Frankfort, KY is certified under EPA for testing Drinking Water & Dairy Water samples.

KPHL - EPA certification does not extend to any other laboratory.

EPA certification requires: annual 10 sample proficiency test with no errors
Intense on-site evaluation (analysts, methods, quality control, etc.)

Note:

There is only 1 State Laboratory in KY – it is in Frankfort, KY

Other water testing labs exist. They may not be EPA certified and their submission requirements, forms, collection kits, rejection criteria, analysis details, and results reporting may be different.
KENTUCKY PUBLIC HEALTH LAB
WATER TESTING

KPHL TESTS THE FOLLOWING:

• PRIVATE DRINKING WATER

• RECREATIONAL WATER
At KPHL we test Drinking Water samples from private sources

WELLS

CISTERNS

SPRINGS

(WHEN USED AS DRINKING WATER)
Private Drinking Water Sources

We do NOT test samples from open springs or other outdoor unprotected bodies of water.....or ponds...... or puddles

The reason.........Testing these kinds of samples with our Drinking Water method would not be meaningful, because this water would be expected to have bacteria, including Coliforms and possibly E. coli.
PRIVATE DRINKING WATER

WHO COLLECTS:

Must be a Registered Environmentalist/Sanitarian or authorized collector

WHAT IS BEING DETERMINED:

Sanitary Quality & Suitability for General Use

TESTING:

Total Coliforms (fecal & nonfecal Coliforms) plus E. coli
Coliforms and E. coli are *indicator organisms* used to assess the safety of drinking water supplies.

The presence of Coliforms in water does not necessarily indicate fecal contamination, but indicates the *possibility* of fecal contamination.

The presence of E. coli in a water sample indicates the *probability* of fecal contamination.
Water Collection Kit Contents:

- Instruction sheet

- Sterile plastic sample bottle with 100 mL fill line etched onto it. (Do not substitute)

- Plastic Ziploc seal bag

- Lab 507 Form

- Styrofoam mailer

- mailing label
Water Collection Kit Contents:

1. Water must be collected in sterile vials provided by Laboratory Services. Plastic vials intended for the collection of water samples contain a dechlorinating agent, sodium thiosulfate. The purpose of sodium thiosulfate is to neutralize residual chlorine in the moment the sample is taken. The thiosulfate prevents continuing action of the bactericidal aims of chlorine during the time the sample is in transit to the laboratory. The bacteriological examination will then indicate more closely the true quality of the water at the time the sample was collected.

2. When sampling from a tap, the following steps should be taken:
   a. Select a tap or hose bibb which has been in use and does not leak. Do not sample from a drinking fountain or gate valve. Remove all attachments from sample tap prior to sampling.
   b. Flush tap for 2-5 minutes before collecting sample. Do not flush tap if source of contamination is suspected to be within the lines of the sampling site.
   c. Identify sample source on sample form and plastic vial.
   d. Fill plastic vial to 100 ml fill line.
   e. Re-cap the plastic vial securely.
   f. Check plastic vial for leaks by shaking and inverting plastic vial several times. Place vial in plastic bag.
   g. Place plastic vial and completed water sample form in the Styrofoam mailer.
   h. Remove backing from mailing label and seal Styrofoam mailer along the short axis.
   i. Pool and health club spa samples require three sample plastic vials each. These can be sent along with the complete form in an appropriate size box to lessen the cost of postage. Beach samples require only one vial.
   j. Samples must arrive in the laboratory within 30 hours of collection. Samples requiring chain of custody precautions are to be fielded and taken to the nearest certified laboratory within 6 hours of collection.

3. A complete and accurate laboratory form must accompany each sample submitted for examination. Indicate if sample is other than drinking water (pool, beach, etc.)
Water Collection Kit Contents:

- ZIPLOC PLASTIC BAG
- MAILING LABEL
- PLASTIC SAMPLING BOTTLE (etched 100 ml fill line)
- STYROFOAM BOX
Water Sample - Collection Kit

Water Collection Kit Contents:

Lab 507 Form
Drinking Water – Collection Don’ts

- Do not collect water from semi-public or public water supplies
- Do not collect water from a “frost-free” hose bib or gate valve
- Do not collect water from a drinking fountain
- Do not collect water from a swinging or swivel faucet
Drinking Water – Preparation for Sample Collection

WASH HANDS

BEFORE COLLECTION

SAMPLES

Note:
Samples submitted to the state Water Lab for analysis must be collected by an “authorized” collector.
Drinking Water – Faucet Hardware

Remove screens, aerators, filters, hoses, or other faucet hardware from the end of the tap. (can harbor bacteria)

TIP: Ask the homeowner to remove the hardware themselves........as damage may occur if the hardware is deteriorated or hard to remove.
Drinking Water - Sample Collection

- Open the cold water tap and flush for 2-5 minutes to clear the service line.
- Do not flush tap if source of contamination is suspected to be within the lines of the sampling site.
- Adjust the water flow to about the width of a pencil and at a steady rate of flow.
- Don’t change the rate of flow once you have started sampling, because this could dislodge bacteria or debris inside the line.
Drinking Water - Sample Collection

- Remove the bottle cap Carefully
  Do not touch the inside of the cap or the bottle.

- Hold the bottle in one hand and the cap in the other.

- Position the bottle under the water flow.

- Fill the bottle to the marked 100 mL fill line.

- Place the cap back on the bottle and close securely.

NOTE:
The “white powder” in the sample bottle is the chemical sodium thiosulfate, which neutralizes residual chlorine that may be in the water.
Drinking Water - Sample Collection

Sample bottles must be filled to the 100 mL fill line.
Drinking Water - Sample collection

If slightly over filled, the sample volume must be adjusted in the lab with a sterile pipette after mixing.

If the sample bottle is over filled all the way to the top, the sample cannot be properly mixed, and removing a portion may cause results to be inaccurate.
Samples submitted to the State Water lab for analysis must be collected by an authorized collector & kept under his/her control until released to a delivery service such as the U. S. Mail......... U P S.......... FedEx......... or other.
WATER COLLECTION ISSUES

- More than 90% of unsatisfactory water samples are rejected due to TIMING issues.

- Samples received >48 HOURS from collection cannot be tested.

- Samples received 30-48 hours from collection will be tested, but a QUALIFYING statement will be added to results stating that results may not be valid.

- If mailing samples through the regular mail from your area does not get them here in one day, please try Priority Mail or an overnight delivery service.
Drinking Water Analysis

1. Colilert media is added to 100 mL of sample

2. Sample poured into Quantitray

3. Culture incubated at 35° C 24 hours

Results are reported after 24 hours incubation
Drinking Water Analysis

Yellow color indicates PRESENCE (growth) of Coliforms
Sample is Total Coliform positive

Fluorescence indicates PRESENCE (growth) of E. coli
Sample is E. coli positive
# Most Probable Number

## MPN Table

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- 0 + wells = <1 MPN/100 mL
- 20 + wells = 25.4 MPN/100 mL
- 36 + wells = 62.4 MPN/100 mL
Recreational Water

KPHL TESTS WATER FROM:

Public Swimming Beaches

Public Swimming Pools
Recreational Water

KPHL TESTS WATER FROM:

WHIRL POOLS

&

THERAPEUTIC POOLS

&

PUBLIC SPAS
Recreational Water Analysis

Swimming Beach and Swimming Pool

Most Probable Number of Total Coliforms and E. coli are reported.
Recreational Water Analysis

Therapeutic Pool samples
Hospital Pools, Whirl Pools, or public spas

Most Probable Number of Total Coliforms, E. coli and Pseudomonas are reported.
Recreational Water

Public Swimming Beaches
**Public Swimming Pools
  1 bottle = 1 set of results
  Total Coliforms – E. coli

**Therapeutic Pools, Whirlpools
  2 bottles = 1 set of results
  Total Coliforms - E. coli - Pseudomonas

**NOTE: These instructions differ from the handbook, because the testing protocol was changed in late 2006. Indicator organisms now being tested for require less sample. If more is sent than required.....no problem 😊.
Sample Submission – Form 507

Authorized collector
Sanitarian number (San.No.)
Collection Date
Collection Time
Owner / Occupant
County
Submitter
LHN site# for environmental office
Indicate if other than a drinking water sample.
Rejection of Water Samples

♦ Sample received > 48 hours from time of collection
♦ No authorized collector given
♦ No date of collection given
♦ Insufficient quantity (less than 100 mL)
♦ Sample bottle broken or leaking
♦ Test not available
Collect and mail drinking water samples on Monday, Tuesday, and Wednesday

Monday and Tuesday for Whirlpool / Spa water samples

Use the best means of delivery to ensure water samples are received in under 30 hours from the time of collection for accurate and valid results.

If mailing samples through the regular mail from your area does not guarantee arrival to the state Water Lab in one day, try Priority Mail or an overnight delivery service.
The Field Handbook for Registered Sanitarians contains collection information for Drinking Water samples and Recreational Water samples.

http://www.chfs.ky.gov/dph/sanitarians.htm
Reporting

Results are sent electronically

*Local Health Network (LHN)*

It is very important that the *submitter number* for your office be stated on the request form
Water Testing Results - Turn Around Time

Water Testing Reports go out overnight & are transmitted the next morning over the LHN to the SUBMITTER location provided on Lab Form 507

- Drinking Water, Public Beach, and Public Pool results should be accessible within 48 hours from receipt in the laboratory

- Results on Therapeutic Pools and Spas tested for Pseudomonas (in addition to Total Coliforms & E.coli) may take longer to receive:
  - NEGATIVE Results - accessible on the morning after test completion (48 hours)
  - Presumptive POSITIVE & Confirmed POSITIVE results may require 4 days
Problems

If there are problems retrieving results, contact:

Custom Data Processing

(502) 695-1999
Waterborne Pathogen Testing

• The Kentucky Public Health Laboratory offers a wide variety of services in the interest of public health, but KPHL cannot do everything.

• Waterborne Pathogen testing will be considered on a “case by case” basis.

• In some instances, it will be more appropriate for this sort of testing to be done at reference laboratories or commercial laboratories.
• Perform a thorough investigation & interview with workers, managers, shippers, consumers, etc. to narrow down and be as specific as possible when requesting a chemical analysis.
  - There is no one “fits all” chemical screen or test.
  - Chemistry equipment and analysis is sensitive, expensive and time consuming

• Back track from the consumer or complainant to the source. Ask questions to possible exposures and activities that involve cleaning, construction, and spraying of chemicals

• Be specific in requesting Herbicide VS Pesticide screening based upon investigation and interviews
AVOID Wrapping Food in PLASTIC!

PLASTIC INTERFERES with the sensitive analytical chemistry instruments.

Exception:
leave the plastic wrap on suspect samples that are already enclosed in plastic, such as lettuce.

Alternative:
Aluminum foil or paper can be used as a wrap (make sure there is no leakage) and then if necessary, it can be placed in a baggie.
CHEMICAL ANALYSIS of FOOD & WATER

Collection Containers

• Collection containers (bottles, jars, etc.) **cannot** be plastic

• Use the glass container in the food kit - approved for chemical analysis

• Do **NOT** use the 100ml bottles in the water collection kit for chemical analysis. They are “plastic” & contain a “dechlorinating” agent
CHEMICAL ANALYSIS of FOOD & WATER

ALTERNATE FOOD COLLECTION CONTAINERS

If containers must be used that are not from the food kit:

- CANNOT BE PLASTIC

- Must be Washed with Soap and Water and Rinsed with Acetone (removes organics)

- A piece of Foil must also be placed between the lid and container if the lid is lined or coated with plastic. Make sure the food or beverage does not come in contact with the lid. Secure the lid tightly so no leakage occurs.

NOTE: Inspect lids in food kits for lids with plastic coating and lining. Older Food kits before March 2006 may contain glass bottles with plastic coated/lined lids.
CHEMICAL ANALYSIS of FOOD & WATER

- **LIQUIDS:** COMPLETELY FILL Container (TO THE TOP)
  Seal and confirm NO AIR bubble is present so volatiles (possible suspect chemical) will not escape

- **DRINKING WATER** takes a minimum of 1 liter; (chemistry will supply containers upon request)
CHEMICAL ANALYSIS of FOOD & WATER

- One 504 Form per sample & control (blank)
- One State Seal per sample & control (blank)
A complete & accurate Laboratory 504 Form must accompany each sample submitted for examination.
A complete & accurate Laboratory 504 Form must accompany each sample submitted for examination.
METALS IN WATER

SAMPLING AND TRANSPORTATION INSTRUCTIONS
FOR THE DETERMINATION OF METALS IN WATER

1. Take sample in the cubetainer provided. It contains Nitric Acid as a preservative.

2. Do Not Rinse Out the cubetainer.

3. IF sample collection is for Lead analysis, take first draw water from the primary drinking water source in household. If you cannot, then let water stand for approximately six hours.

4. Fill out Lab-504 Form provided completely.

5. Ship to Laboratory Services as soon as possible using the following address:

   North Loading Dock
   Instrumentation Chemistry Section
   Division of Laboratory Services
   100 Sower Blvd., Suite 204
   Frankfort, KY 40601
   Attn: Metals in Water

Cubetainer is for METALS testing only
Submit food samples at their appropriate temperature.

Examples:

a. Dry perishables like cold cereal – Room Temperature
b. Dairy Products – Refrigerator temperature
c. Ice cream – Freezer Temperature

Do not package & ship foods in “loose” ice, use cold packs or “blue” ice
CHEMICAL ANALYSIS – QUESTIONS??

CONTACTS:

PESTICIDE LABORATORY
(502) 564-4446/ EXT 4409
IRA FINK - ira.fink@ky.gov

CHEMISTRY SUPERVISOR
(502) 564-4446/ EXT 4439
ISABELLE BERBERIAN
isabelle.berberian@ky.gov

FAX: 502-564-7019

When in Doubt or Questions ……Call
Special Consideration - Human Origin/Source

The Kentucky DLS Laboratory
Does **NOT**
Analyze Food or Water for
Human Origins or Sources
Such as Tissue or Blood

REFER TO THE FORENSIC LAB
(502) 564-5230
(Forensic Biology Caseworking Unit)
SAMPLE COLLECTION
FORENSIC LAB - GUIDELINES

• Wear Clean Disposable Gloves

• Instruments used to Collect & Transfer Food item(s) should be Clean

• Collection Containers should be Sturdy, Leak Proof, and Clean (avoid containers that encourage bacteria, mold, or mildew).
  1. Paper Wraps, White Typing Paper, or Butcher Paper
  2. Wax Paper, Lab Collection Containers
  3. Glass Jars are suitable for liquids
• Seal and Appropriately Label

• **Freeze** it as soon as possible.

• Place in a Transport Container

• **Keep Frozen** until it can be submitted to the Forensic Lab for analysis via an appropriate law enforcement agency.

• **Law Enforcement** has necessary paperwork
CENTRAL FORENSIC LAB

CONTACT PERSON:

EDWARD TAYLOR
KENTUCKY STATE POLICE
CENTRAL FORENSIC LAB
(502) 564-5230 Ext# 309

OR

ASK FOR FORENSIC BIOLOGY CASEWORKING UNIT
Contact Local Emergency Management
They are TRAINED to RESPOND

Call Epidemiology (502-564-3418) or 888 – 9 –REPORT (737678)

Call Preparedness Branch (502-564-7243)

Contact Lab Services (502) 564-4446

NOTE: Allows follow your internal communication policy
BIOLOGICAL INCIDENT

 Reported to the Emergency Operation Center (EOC) & assigned an EOC#

 Emergency Operation Center – 502-607-1630
 1-800-255-2587

 Required Documentation:
 1. Request for Examination
 2. Chain of Custody
 3. Results & Sample Acquisition/ Destruction

 Forms on line - www.chfs.ky.gov/DPH/info/lab
ENVIRONMENTAL (BT) CHECKLIST

TRIAGE:

- Chemical
- Radiological
- Explosives
- Biological

Submit a Copy of Results

SIZE:

8”H x 10”W x 15”L
(maximum limit)

PACKAGING:

- Double - bagged/contained
- Exclude gloves/cleaning materials
- Outer packaging decontaminated
  (i.e. 10% bleach soln)

TRACKING:

EOC# (Call EOC: 800-255-2587
502-697-1630)

FORMS:

- Request for Examination
- Chain of Custody
- Results Acquisition
forms: http://chfs.ky.gov/dph/info/lab/

CONTACTS:

- Lab Services
  502-564-4446 (Mon-Fri: 8am-4:30pm)
  502-330-6235 – After hours paging
REQUEST FOR EXAMINATION FORM

LABORATORY SUBMISSION FORM COMPLETED by the SUBMITTING PERSON/AGENCY
Samples must be accounted for at all times from the point of collection to the submission of the sample to the State Laboratory.

**SUBMITTER COMPLETES FORM WITH INFORMATION (THOROUGH)**

(If a sample is submitted with a Law enforcement chain of custody, the Submitter additionally completes this form)

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### Chain of Custody Form

<table>
<thead>
<tr>
<th>ITEM NO.</th>
<th>DATE</th>
<th>RELEASED BY</th>
<th>RECEIVED BY</th>
<th>PURPOSE OF CHANGE OF CUSTODY</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**ALL SIGNATURES are REQUIRED**
FOOD & WATER
collection, packaging, testing, & communication

COLLABORATIVE EFFORT
OF
PUBLIC HEALTH
PROFESSIONALS

THANK YOU !!!!!!
Cabinet for Health and Family Services

CONTACT INFORMATION

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