Colorado Department
of Public Health
and Environment

First Responder Manual
On
All-Hazard Environmental Incidents
Technical Support & Sampling

Colorado Department of Public Health & Environment

2008 Edition

Colorado Department Public Health and Environment (CDPHE)

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National Response Center (24-hour)	800-424-8802
Local Emergency Planning Commission	303-273-1622
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INTRODUCTION

Occasionally both technical support and laboratory diagnostics are required to assist the on-scene commander in the decision process when an unknown or suspicious material is involved in an incident. The Colorado Department of Public Health and Environment (CDPHE) provides technical support to community first responders, other state agencies and federal agencies for incidents involving biological, chemical and radiological agents.

Laboratory diagnostics are available for incidents involving criminal acts (i.e. violators of hazardous materials regulations or laws) or credible threats and incidents impacting the public's health. Specific steps must occur before samples can be taken to CDPHE's laboratories for both the protection and safety of laboratory personnel as well as ensuring limited resources are used wisely and the laboratory is prepared to receive the samples.

Purpose

This document is to serve as a resource to first responders. The goal is to provide first responders with information and guidance related to technical support and environmental sample collection expectations. It will detail procedures for sample collection and submission to the CDPHE's Laboratory Services Division. The technical support provided by CDPHE can occur through multiple divisions based on the combined information shared by the on-scene responders through their hazard assessment and the on-scene safety assessment of the impact on first responders or the public.

Process

The objectives, scope and content of this document were developed through a series of workgroup meetings comprised of state and local public health professionals, state and local hazardous materials specialists, U.S. Postal Service Inspection Services, and the Federal Bureau of Investigation (FBI). The 2008 updates to the manual also included the Colorado National Guard Civil Support Team.

Reporting Events

To report an emergency event, a hazardous substance spill or request immediate technical support, contact CDPHE's Emergency Response Line at 1-877-518-5608. CDPHE's Emergency Response Line is staffed 24 hours a day, 7 days a week by the department's Emergency Preparedness and Response Division. This division will coordinate the appropriate expertise for each situation, including access to the department's Laboratory Services Division during non-business hours for credible threats.* **No sample will be accepted at the CDPHE Lab without prior notification AND approval by this department.**

*Note: CDPHE is not a first responder agency. The department provides technical expertise as secondary responders supporting the on-scene responders. CDPHE does not retrieve or provide transport for samples intended to go to the department's laboratory. The transport of samples is the responsibility of the on-scene agencies.

A. Requesting Technical or Laboratory Support of CDPHE

Once an incident is reported to CDPHE, assistance can be requested. The following will subsequently occur for each type of incident:

General Emergencies – Technical Support

The Emergency Preparedness and Response Division will notify:

- Local public/environmental health departments in the jurisdiction of the incident
- CDPHE Divisions assisting (based on the potential of biological, chemical or radiological agents involving air, water, food or soil). This may include:
  - Air Pollution Control Division (for air modeling and asbestos)
  - Consumer Protection Division (for food, dairy and consumer products)
  - Disease Control and Environmental Epidemiology Division (disease investigation, control)
  - Hazardous Materials and Waste Mgmt Division (chemical, radiological, waste)
  - Water Quality Control Division (drinking and waste water)
- Appropriate other entities including federal agencies such as FDA, USDA, EPA and DOE

Credible Threats – Technical Support *(see Credible Threat Section for details)*

The Emergency Preparedness and Response Division will notify:

- Local public/environmental health departments in the jurisdiction of the incident
- CDPHE Laboratory Division and other divisions assisting (based on the potential of biological, chemical or radiological agents involving air, water, food or soil)
- Colorado Division of Emergency Management (verification call)
- Colorado Department Public Safety, Office of Preparedness and Security
  Federal agencies as appropriate (e.g. FBI, EPA, HHS, DOE)

General Emergencies or Credible Threats – Laboratory Diagnostics

The Emergency Preparedness and Response Division will notify:

- CDPHE Laboratory Services and other divisions assisting (based on the potential of biological, chemical or radiological agents involving air, water, food or soil); the laboratory may be contacted directly at 303-692-3090 or pager: 877-705-1016

FIELD OPERATIONS

A. Scene Characterization

Scene characterization is a concise summary of information from the scene that is quickly assembled (within two hours). When responders are seeking technical and laboratory support, this initial field assessment should occur prior to contacting CDPHE. At the time CDPHE is contacted the following information should be relayed as the Scene Characterization Report:

1. Hazard Assessment/Immediate Risks
   (a) Identify if the incident is 'Accidental' or 'Deliberate'
   (b) If deliberate, determine if it's a 'Credible Threat' or 'Non-Credible Threat'
   (c) Identify type agent and media (e.g. biological, chemical, radiological; air, water, soil, food)
2. Safety Assessment
3. Rapid Field-Test Results
4. Documentation/inventory of samples

From this information a sample collection process is developed by the Laboratory Services Division to support on-scene response and the chain-of-custody requirements for legal action. Ideally, standard formats should be used for the Scene Characterization Report, specifically the field tests and the sample collection documentation (described in greater detail below).

1. Scene Characterization Report - Hazard Assessment Section

Since the events at the scene are typically still unfolding when the Scene Characterization Report is initially generated, the Hazard Assessment section of the report can be brief and added to as the details unfold.

a. Hazard Assessment Section -Accidental/Deliberate/Suspicious

It is important to determine if the incident is accidental or deliberate. Items are classified as suspicious when:
(1) Unknown substance exists (including those in packages or envelopes)
(2) Threatening communication is associated with the item
(3) Illness is associated with the item

When there is an unknown substance release with no threat or illness associated a logical explanation for the substance's presence must be ruled out. If an envelope or package exists, often the name of the company sending the item can provide assistance in obtaining an explanation. If a reasonable and defendable explanation occurs, then the item is determined to be a 'non-credible' threat. Further action and support from CDPHE will be based on the potential risk to the public's health or the environment.

** Note: CDPHE will not test samples collected from 'non-credible' threats except under a 'fee-for-service' basis and with prior laboratory approval.

b. Hazard Assessment Section - Deliberate; Credible/ Non-Credible Threat
Any situation, accidental or deliberate, that is a violation of hazardous materials regulations or is a criminal act requires specific steps occur to support legal action. This needs to be determined early to ensure chain of custody occurs.

The threat assessment and credible/non-credible threat determination is based on information received by responders at the scene. If the threat is deemed 'credible' specific steps are taken to protect the public and the responders and this should be communicated to the appropriate officials.

- UNOPENED Letter/Package Threat Assessment:
  - Oily stains or suspicious discolorations or powder on package
  - No Return Address, Foreign, fictitious address
  - Foreign mail, airmail and special delivery
  - Restrictive markings, such as confidential, personal, etc.
  - Excessive postage stamps
  - Handwritten or poorly typed addresses
  - Incorrect titles
  - Titles, but no names
  - Misspelling of common words
  - No return address
  - Excessive weight
  - Rigid envelope
  - Lopsided or uneven envelope
  - Protruding wires or tinfoil
  - Excessive security materials, such as masking tape, string, etc.
  - Visual distractions

- OPENED Letter/Package Threat Assessment:
  - Liquid, spray, powder or vapor
  - Unusual odor
  - Threatening notes

If the suspicion of a threat cannot be resolved, notify immediate supervisor, police, fire and the FBI. For credible threats notify:
- Immediate Supervisor
- **Local police, fire and hazmat**
- County and State emergency manager
- FBI\ Local, county and state health departments

If individuals are potentially exposed, a decision must be made in a reasonable period of time (preferably less than 2 hours) to:

- Release the individuals without decontamination or follow-up
  Release without decontamination but public health or medical follow up
  Decontaminate and then release individuals without follow-up
  Decontaminate and then release with public health or medical follow-up

*Obtain the names and contact information for each individual potentially exposed so that public health or medical follow-up can occur. Turn over the information to the local public health agency or medical support personnel on-scene.

c. Hazard Assessment Section -Type of Agent

DOT papers, signage, company experts, and responder experience is all beneficial in determining the type of agent. It is important for responders to summarize the methods used to obtain this preliminary information. The type of agent should include preliminary identification of the:

(1) Agent classification as biological (including toxins), chemical or radiological
(2) Media type as powder, liquid, vapor that exists in the soil, water, air or food
(3) Quantity of material (best estimate)

2. Scene Characterization Report – Safety Assessment Section

The dangers associated with an agent are influenced by the use of personal protective equipment, weather conditions and other factors. This ultimately becomes critical to the safety of the responders and the public's health. For suspicious envelopes/packages or shipments vehicles an attempt to contact the sender of the envelopes/package to verify contents may assist in determining risk or threat status. Responders should identify all persons who touched the item to assist in surveillance, medical monitoring or implementation of health and safety measures.

a. Safety Assessment - Credible Threat and Envelopes/Packages
Safety for UNOPENED Envelopes/Package – Credible Threat:

- Wash hands with soap and water if the item was touched
  Double wrap the item in plastic wearing gloves (latex, nitrile or vinyl), a particulate mask and move to a secure location if its not leaking
- Treat as crime scene and proceed as with opened letters/packages

Safety for OPENED Envelope/Package – Credible Threat:

- Do not touch, smell or inhale near the item and substance
- Avoid hand contact with outer clothing or skin surfaces
- Keep mouth and nose closed or cover face with sheets of paper or protective mask
- Evacuate persons from at-risk areas to minimize potential exposure
- Isolate area and deny entry
- Wash hands with soap and water if the item was touched
- If clothing is contaminated, remove outer clothing, place in garbage bags and label the bag 'BIOHAZARD' (one bag per person)
- Give clothes to law enforcement for lab analysis
- Treat as a crime scene if suspicion cannot be resolved or a threat is received

Note: Approximately 80 percent of contamination can be removed by taking off outer garments

All samples must be screened for volatile organic compounds, explosives, incendiaries, and ionizing radiation prior to sample collection. Present a copy of this document to the laboratory at the time the sample is delivered.

b. Safety Assessment - Credible Threat Sample Transport

Safety measures must include assessing the package or material for potential secondary devices, particularly explosives, prior to transport of samples to the CDPHE laboratory. The FBI and local law enforcement are expected to provide CDPHE with confirmation that such safety checks and radiological screen occurred prior to the sample leaving the scene. Be prepared to provide the laboratory information on the packaging and labeling of shipping containers, the transport vehicle and the identification of the driver. CDPHE will provide driving instructions to the correct laboratory facility and any safety criteria essential to accept the sample. **Sending samples to CDPHE laboratories for testing must be pre-approved by CDPHE.**

3. Scene Characterization Report – Rapid Field Testing

a. Field Testing Process

Field testing is the process of screening an unknown material to obtain a preliminary identification of the materials as a 'toxic industrial chemical' (TIC), biological agent, potential WMD or other agent. The objective of field screening is to:

- Tentatively identify the contaminant (definitive laboratory testing to occur later)
- Rule out potential risks (for hazard reduction and mitigation), which include:
  - Explosives/ volatility
  - Radiological agents
  - Chemical agents
  - Flammability/ volatility
  - Corrosive/pH determination
- Determine if isolation, evacuation or other measures are warranted
- Initiate chain-of-custody steps for formal samples

If the material is of a small enough quantity that the entire sample could be consumed during field testing, retain the sample for more definitive laboratory testing and do not destroy the entire sample by field testing (but a non-destructive field test may be used).

Tests should be performed with multiple testing methods when possible. All field screenings and sample collection steps should be performed in accordance with nationally recognized standards. Appropriate personal protective equipment (PPE) should be worn. It is important to also collect and test background samples in the area of the contaminant to compare the test results. When possible photograph sample sites prior to sampling in accordance with local law enforcement procedures and practices.

On-site responder testing equipment utilized for the field screening tests may include, but is not limited to:

- colorimetric tubes
  biological immunoassay "tickets"
  military type paper chemical agent detectors (M8 paper)
  military type wet chemistry chemical agent testing detectors (M256 kits)
- chemical agent monitors
- surface acoustic wave (SAW) monitors
- infrared spectrometers
- chemistry categorization kits

The accuracy of these tests varies and false positives, misinterpretation of results and cross sensitivities with non-hazardous substances can occur. If a field screening is falsely reported as positive, the result could provoke unnecessary concern, evacuations, panic, and media attention. Action taken on a false positive could damage the credibility of the agency and its personnel. Likewise, a negative result on a field-screening test for a true hazardous agent could be more damaging. Therefore, field test results should be referred to as 'presumptive' positive or negative.

b. Field Testing Support- National Guard

The Colorado National Guard 8th Civil Support Team (CST), Weapons of Mass Destruction Unit, works for State Governors under the command and control of the Adjutant General. The CST can deploy to a scene and link up with the civilian incident commander to provide a direct-support military relationship but CST always remains under military control. The team can provide support in the form of conducting slightly more advanced field testing than the typical local hazardous materials response team.

Requesting operational deployment of the 8th Civil Support Team is accomplished through contacting the State Emergency Operations Center (EOC) at their 24 hour number 303-279-8855. For general assistance contact the 8th CST-WMD through the 24 hour Staff Duty Officer at 720-847-6874. The anticipated deployment time is 4 hours from a validated alert; they do strive to have the team out the door within 2 hours.

c. State Health Dept Laboratory Support
If samples are desired to be tested by CDPHE or confirm on-scene preliminary analysis, a scene-specific sample collection plan must be developed. CDPHE's Laboratory Services Division and other entities involved on-scene will utilize the Scene Characterization Report to assist in developing the sample collection steps. A briefing among relevant parties is recommended to ensure that the transition from the scene to laboratory analysis occurs smoothly. **Samples will not be accepted at the CDPHE lab if laboratory notification and approval does not occur prior to sample delivery.**

When a sample is forwarded to the CDPHE lab for additional laboratory testing include: a list of the performed field screening tests and their results; the manner in which the samples were collected; the quantity collected per sample.

4. Scene Characterization Report – Documentation

a. Documentation – Inventory of Samples
A summary document should be created that lists the location and sampling method for each sample collected. Use the sample identification number as the key identifier for each sample listing. Ideally a copy of this summary document should accompany the samples to the laboratory as the information may be important for the laboratory techniques.

Save items such as immunoassay "tickets" and buffer solutions for possible future legal action related to the material. When chemical or biological monitors have a "data logging" capability or a printout capability, save or record the data as well.

b. Documentation – Record Maintenance
It is important to maintain records of all scene characterization activities, even for potential threat incidents that were ultimately dismissed as 'not credible.' Documentation about a particular activity can be accessed long after the details of the incident have faded from memory.

B. Human Exposure

It is critical a decision be made in a reasonable period of time (preferably less that two hours after exposure) as to the action to take for individuals potentially exposed to the substance. If the level of hazard is not known, professional judgment should be used to ensure these individuals receive proper medical care, if needed.

It is important that law enforcement is informed of the decision pertaining to human exposure for any incident involving a credible threat; proper handling of items deemed evidence is important.

Any person believed contaminated with a hazardous substance and displaying symptoms should be decontaminated prior to leaving the scene. Proper procedures should exist to ensure privacy and dignity is maintained. Proper clothing should be available for each person after decontamination is complete.

The name(s) and contact information for all individuals potentially exposed should be recorded and the local public health department contacted so surveillance for potential illness can occur.

EMS should be properly briefed prior to transporting any individual requiring a medical assessment. No individual that is contaminated should leave the scene in an ambulance without prior approval from the ambulance agency. Appropriate PPE should be worn if a contaminated person is being transported. Any ambulance that is contaminated must be properly cleaned prior to re-use of the vehicle.

C. CDPHE Laboratory Use

Although there are other laboratories in the state, this section will pertain only to the CDPHE laboratory services. The CDPHE's Laboratory Services Division functions as the state's principal public health and environmental laboratory. The mission of the Laboratory Services Division is to protect the health and environment of all Coloradoans from infectious and metabolic diseases, environmental pollutants, and acts of terrorism by providing accurate and timely laboratory analyses and information. Specific details about the services the CDPHE laboratory can be found at the division's homepage: http://www.cdphe.state.co.us/lr/index.htm

CDPHE accepts human specimens (blood, sputum, aspirates, etc.), fomites (documents, powders, food, soil, etc.), and culture isolates (identification/confirmation). Laboratory testing for biological, chemical and radiological agents can occur, based on the type of sample.

CDPHE Lab Use and Threat Credibility
Based on the credibility of the threat, the following assumptions exist for laboratory diagnostics:

1. **Non-Credible Threat Laboratory Diagnostics**
   No testing is warranted. Public Health officials may receive demands for testing and may offer fee-for-service testing options including powder identification or rule-out cultures for Bacillus anthraces or other agent. If testing is desired but rejected by CDPHE, responders should consider having desired tests performed at private laboratories or qualified local public health.

2. **Credible Threat Laboratory Diagnostics**
   Testing is warranted. FBI is responsible for the investigation of credible threats; CDPHE's laboratory must receive confirmation from the FBI before acceptance of any sample related to a credible threat occurs.

Public Health officials are responsible for intervention and communication pertaining to protecting the public's health and notification of the diagnostic findings will occur with local public/environmental health as well as the responding agency and the FBI.

Prioritizing Samples and Tests

CDPHE's laboratory will prioritize samples based on public health incidents and credible threat status. The Credible Threat Specimen Triage Guide (Chart 1.0) below is utilized to determine threat-related prioritization. The responder will be informed by laboratory personnel of the priority status for their samples at the time of the testing request. Those submitting samples should follow the protocols outlined in the **Sample Collection** section to ensure samples submitted are processed in a timely manner and provide the best results feasible.

Chart 1.0: Credible Threat Laboratory Specimen Triage Guide

Human Specimens / Culture Isolates	Environmental Specimens
Category 1- HIGH PRIORITY	
From cases with illness - signs and symptoms were reviewed by a clinician or epidemiologist as consistent with infection/intoxication due to a recognized biologic, chemical or radiologic (CBR) agent associated with a credible threat.	Collected from a scene or associated with an event that is deemed a credible threat for a CBR agent by the FBI, in concert with local law enforcement.
Action: Perform testing immediately on receipt of specimen (24/7) and refer to other federal or state laboratories as soon as possible. **Testing**: *Conduct testing with multiple methods*	
Category 2- Intermediate Priority	
From cases with illness that is a low possibility of causation by a CBR agent (i.e. the patient is ill but the clinical picture is not typical of a CBR agent).	Collected from a scene or associated with a criminal act (felony menacing, hoax) with a specific CBR agent known, but no credible threat known.
Action: Perform testing on the next regular business day **Testing**: *Perform testing using a single method for the threat agent on probable criminal specimens; other testing deemed appropriate as indicated by the situation*	
Category 3-Low Priority	
From patients with no discernible illness – or – specimen was obtained for epidemiologic studies.	Collected from scene that was deemed a non-credible threat.
Action: Refer submitters to local health department or private laboratories offering testing for threat agents or for the identification of a substance on a fee-for-service basis. * **Testing:** *As requested by submitter*	

Environmental testing for anthrax (1) and powder identification (2) is available on a fee-for-service basis by:

- Industrial Laboratories, 1450 East 62nd Avenue, Denver, CO 303-287-9691 (1, 2) or
- Colorado State University Veterinary Diagnostic Laboratory, Ft Collins, CO 80523 970-491-1281 (1)

SAMPLE COLLECTION

A. General

Collect general-purpose samples only after law enforcement, hazardous materials, and public health personnel have determined if there is a credible threat or legal action required related to the incident or substance. Properly trained HAZMAT personnel using appropriate personal protective equipment as specified in OSHA standard 1910.120 should collect samples. Duplicate samples may need to be collected for law enforcement/evidence purposes.

NOTE: Samples to be submitted to the CDPHE laboratories must be pre-approved and properly packaged. CDPHE will provide specimen and packing guidelines.

B. Chain-of-Custody

Follow the appropriate security measures for samples considered evidence. Initiate Chain-of-Custody steps prior to sample collection and keep samples under the control of designated personnel at all times. When samples are not in the possession of the designated personnel, secure samples (e.g. locked in a secure area). Closely track and document the Chain-of-Custody steps for each sample, using a 'Chain-of-Custody Form.'

Steps for Sample Collection

1. Change gloves between each sample to prevent cross-contamination
2. Collect no more than 10 grams of material per sample
3. Place each sample in an unused, self-sealing sterile bag
4. Properly label each bag (see below) after sample is placed in the bag, seal the bag
5. Clean the outside of the sealed bag with sodium hypochlorite (concentration = 0.5-0.6%) just prior to leaving the contaminated area
6. Place the cleaned sealed bag in another unused self-sealing bag
7. Place contaminated discarded items in a biohazard bag; proceed to decontamination area
8. Place the bag in a shipping container *(See Packaging and Shipping section for details)*.
9. Transport samples at ambient temperature, unless otherwise indicated, to the CDPHE

The CDPHE Lab will NOT accept items with dimensions exceeding 11 ½ inches in size

For Chain-of-Custody samples document on the label the following:

- Unique number or identifier for each sample
- Type of analyses requested (biological, chemical, radiological – specific test)
- Sample location description
- Type of sample (liquid, solid – object or powder, food, etc)
- Date and time of sample collection
- Name of person collecting sample
- Optional - Map of the sample area (consider photographing the collection site)

C. General Information

Consider obtaining technical support from a local public or environmental health agency, or an appropriate CDPHE program, prior to sampling. The goal of chemical and biological sample collection is to detect and characterize the presence of a contaminant. A biological/microbial assessment can occur from analysis of building or environmental materials such as carpet, office equipment, supplies, vials of dust, mail, clothing, heating, ventilation and air conditioning (HVAC) filters etc. To collect these bulk samples, follow the appropriate sample collection method listed below.

Prepare sample labels for each container before beginning sample collection (to minimize the time spent on the site during the sample collection stage). Transfer the information captured on the sample labels to the Chain-of-Custody Form, which can also serve as the sample inventory. See the *'Packaging and Shipping'* section for packaging, transport and shipping guidance.

The CDPHE Lab will <u>not</u> accept items with dimensions exceeding 11 ½ inches in size

<u>General Materials for All Sample Collections:</u>

- Non-powdered, sterile gloves (such latex, nitrile, or vinyl gloves)
- Dacron swab (not cotton or calcium alginate)
- Non-sterile self-sealing bags
- Permanent markers
- Labels and forms – for sample site mapping of the scene
- Sodium hypochlorite (0.5% – 0.6% concentration) in a wash or squirt bottle
- Shipping container approved for transport (See ***Packaging and Shipping*** section)
- Biohazard bag for discarding contaminated materials
- Chain of Custody Form – Required for all credible threat samples

Items that might be useful:

- Sample Documentation Form (*serves as the sample inventory tracking form*)
- Ultra Filtration Field Concentration Apparatus
- Camera (*for documentation of sample collection sites*)
- Sealable Plastic Bag (*bubble wrap baggies can be used*)
- Shipping Container or Rigid Shipping Container

It is important to follow any special laboratory requirements regarding sample collection and transport as this may affect the quality of the analytical results.

Verify that any hatches, locks, etc. are properly secured before leaving the site. Remove all PPE at the site perimeter and place disposable PPE and waste material into a heavy-duty plastic trash bag before leaving the scene. Properly label this contaminated waste.

1. Collecting Surface Samples: Swab/Wipe Method

The swab method is used for biological/microbial sample collection on small, non-porous surfaces that do not have a large accumulation of dust and dirt such as keyboards, hard-to-reach areas within machinery, mail sorters, ventilation grilles, etc.

The wipe method is used for sample collection on large (> 100cm2 or 1 ft^2), non-porous surfaces such as tabletops, counters, desks, file cabinets, windowsills, floors, mailboxes, non-carpeted floors, etc.

Obtain the materials outlined in '*General Materials for all Sample Collections*' (page 16) as well as these additional items:

- Sterile 3" X 3" or smaller synthetic (non-cotton) gauze pad
- Tweezers (if needed)
- **Solution to moisten** swab - Sterile saline (0.85%) or Phosphate buffered saline (PBS) 0.1M, pH 7.2
- Sterile conical centrifuge tube (polypropylene or polystyrene)

Collection Procedure:

1) Don sterile, non-powdered gloves over the standard PPE gloves and clothing
2) Pre-label each container
3) Transfer label information to a Chain-of-Custody Form (see ***Chain-of-Custody*** *section*)
4) Aseptically obtain a sterile 3" X 3" or smaller synthetic (non-cotton) gauze pad
5) Moisten the gauze with sterile saline or sterile phosphate buffered saline (PBS)
6) Wipe the surface being sampled approximately 1 square foot.
   - Avoid letting the gauze pad dry completely
   - Make enough vertical S-strokes to cover the entire sample area
   - Fold the exposed side of the pad
   - Make horizontal S-strokes over the same area
7) Place the sampled gauze in a sterile conical vial, and cap the vial – OR – Place the sampled swab in a sterile conical centrifuge tube, break off the shaft below the area that was held during sampling and cap the tube
8) Ensure vial or tube is labeled and place it in a self-sealing bag
9) Follow Chain of Custody steps

2. Collecting Surface Samples: HEPA Vacuum Method

This method is used for biological/microbial sampling of large porous or non-porous, dusty or dirty surfaces such as carpeting, upper surface of ceiling tiles, ventilation systems, and papers.

Obtain the materials outlined in 'General Materials for all Sample Collections' (page 16) and these items:

- Non-powdered, sterile gloves (such latex, nitrile, or vinyl gloves)
- Dust filter sock (Midwest Filtration Company or equivalent is preferred)*
- HEPA vacuum with collection nozzle
- Rubber bands
  Biohazard bags
- Sterile bags
- Alcohol wipes
- Shipping container approved for transport (see ***Packaging and Shipping*** section)

Note: *If the number of CFUs per gram of dust is desired, use pre-weighed filter socks – or – the mean filter weight of several socks as a background, representative weight.*

Pre-label each container to minimize the time spent on the site collecting samples. Transfer the label information to the Chain-of-Custody Form (see ***Chain-of-Custody*** section)

Collection Procedure:

1) **Don sterile, non-**powdered gloves over the standard PPE gloves and clothing
2) Insert a cone-shaped Dust Collection Filter Sock into the vacuum cleaner nozzle
3) Fold the plastic sleeve over the outside of the nozzle
   - Secure with an elastic band or hold firmly in place using a gloved hand
4) HEPA vacuum the surface: Make one pass of the entire sampling area at a slow rate (12 inches per 5 seconds) Note: *1-2 tablespoons of vacuumed debris are desired*
5) Remove the tape/elastic band; discard as contaminated waste material
6) Remove the cone-shaped dust collection filter sock; place in a self-sealing bag
7) Roll the filter and place it in a sterile conical bag
8) Label the bag; place the cleaned sealed bag in another unused self-sealing bag
9) Clean the outside of the sealed bag with a sodium hypochlorite/bleach solution
   - Use 0.5 to 0.6% concentration of sodium hypochlorite
10) Place contaminated materials into a biohazard bag; proceed to decontamination
11) Place the bag in a container approved for transport

To collect another sample, wipe the nozzle with an alcohol wipe,* change gloves, and repeat steps 1-11. Note: Alcohol wipes will physically remove contamination from the nozzle surface but will not sterilize the surface. *To determine if cross-contamination of samples occurs, occasionally insert a filter sock into the vacuum nozzle after a sample is collected and the nozzle cleaned. Withdraw the sock and place in a sterile conical tube for analysis.*

3. Collecting Credible Threat Samples

Confirmation of a credible threat or other legal action must occur prior to samples being approved and taken to CDPHE's laboratory.

If a credible threat is established and a visible powder or liquid is present on surfaces, collect a swab or wipe sample. Follow proper sampling technique for all other types of samples and desired testing.

If a package or container needs to be opened or disturbed to access visible substance, do NOT open. Instead, submit the item intact to the lab. Follow proper transport steps.

Note: *Avoid submitting clothing, office products, furniture, or other such* ***items unless prior approval for submittal is obtained from CDPHE.***

4. Collecting Water Samples

Technical support may be required through the local public/environmental health agency or CDPHE's Water Quality Control Division. Water samples are accepted for chemical and biological/microbial agent analysis from drinking water, ambient water, and wastewater. Water samples may be from surface sources, storage tanks, pressurized pipes, or other distribution system element (see Figure II-2 for corresponding human tests when samples are related to outbreaks).

Samples from large bodies of water such as reservoirs, whether finished or source water, requires different sampling techniques than those used to collect samples from distribution systems. The EPA Environmental Response Team's standard operation procedure #2013 is an acceptable technique for collecting sampling from these types of water sources.

Obtain the materials outlined in 'General Materials for all Sample Collections' (page 16) and these items:

- Preservative and/or Dechlorinating Agent (if needed)
- Ultra Filtration Field Concentration Apparatus
- Sealable Plastic Bag (bubble wrap baggies can be used)
- Shipping Container or Rigid Shipping Container
- Frozen Ice Packs (preferred) or Sealable Freezer Bags filled with ice
- Heavy Trash Bag

Pre-label each sample container to minimize the time spent on the site collecting samples. Transfer the label information to a Chain-of-Custody Form (see ***Chain-of-Custody*** section).

Procedure – For Water Faucet Samples:

1) Don sterile, non-powdered gloves over standard PPE gloves and clothing
2) Check for in-line filters (home treatment devices) that might interfere with sampling
   - Remove devices, if present; collect sample
   - Collect sample from the device if it cannot be removed; note this on the label
3) If water is collected from a faucet, flush the water tap for a sufficient time to displace the water in the lines (*to obtain a representative sample from water distribution systems*)

Note: Keep the flow rate sufficiently low in order to avoid splashing and aerosolizing water droplets. Divert water to a drain, if possible.

If the water flushed from the tap poses a hazard to the discharge area, consider collecting the discharge for decontamination.

If the decision is made to analyze the samples immediately, contact the lab as soon as possible so they can prepare for sample arrival.

If the decision is made to hold samples (rather than send to the laboratory for immediate analysis), consider the stability of the suspicious agent in unpreserved samples. Preserved sample holding times are based on the respective analytical methods desired and are typically 7-28 days for properly preserved samples. Chill samples, but protect from freezing when holding. If a threat is of concern, be certain to hold samples until the threat evaluation is complete and the decision is made to either analyze the samples or close the investigation.

Procedure – For Chemical Testing of Water:

If the water samples are considered to be hazardous, it may be necessary to implement certain sampling techniques in addition to the guidelines presented below.

For Open-Top caps and septa – seal sample containers; make certain the Teflon (smooth) side is facing towards the water.

For Closed-Top caps (pesticides, etc.) – fill the container to the top, leaving very little or no headspace.

If necessary add a preservatives and/or de-chlorinating agents to collection containers.

Preservatives and/or de-chlorinating agents may be added to the sample containers during sample kit preparation to decrease time of sample collection.

1) Fill sample containers with water flowing from the sample tap
   - Do NOT use rubber or plastic tubing during the collection step
   - Do NOT rinse or overfill the sample containers (for containers with a preservative or de-chlorinating agent)
2) Wipe the outside of the sealed containers with an antiseptic wipe or mild bleach solution
3) Attach a custody seal to the individual sample container (if required)
4) Place the sample container into a sealable plastic bag
   - Bubble wrap baggies can provide protection against breakage of glass containers
5) Place the sealed bags containing the samples into an appropriate rigid shipping container
   - Pack with frozen ice packs (preferred) or sealable freezer bags filled with ice
   - If ice is used, the bag should be thoroughly sealed to avoid leakage
6) Transport samples at ambient temperature, unless otherwise indicated

Procedures – For Microbial Testing of Water

Sampling technique for microbial contaminants may vary based on the agent suspected. The following specific steps should be followed based on the type of testing requested.

Bacteria Tests – Collect a 4-liter sample of water.

Virus Tests – Collect 100 and 1,200 liters of water and filter through a positively charged filter. Send the processed filters to the laboratory; viruses adsorbed to the filter can be eluted in the field and shipped as a one-liter concentrate to another laboratory for further processing.

Protozoa Tests - Collect a 10-liter sample of water (Method 1623: Cryptosporidium and Giardia in Water by Filtration by IMS/FA, EPA-821-R-99-006, April 1999, http://www.epa.gov/nerlcwww/).

Add a preservative and/or de-chlorinating agent (if required) during the sample kit preparation phase to decrease the complexity and time required for sample collection.

1) Fill sample containers with water flowing from the sample tap
   - Do NOT rinse or overfill the sample containers (*Particularly if a preservative or de-chlorinating agent exists in the container*)
2) Wipe the outside of the sealed containers with an aseptic wipe/mild bleach solution
3) Attach custody seal to the each sample container (if required)
4) Place the sample container into a sealable plastic bag
   - Bubble wrap baggies can provide protection against breakage of glass containers
   - Additional instructions for packaging samples potentially containing infectious biological contaminants are provided in the 'Packaging and Shipping' section
5) Place the sealed bags containing the samples into an appropriate, rigid shipping container
   - Pack with frozen ice packs (preferred) or sealable freezer bags filled with ice
   - If ice is used, the bag should be thoroughly sealed to avoid leakage
6) Transport samples at ambient temperature unless otherwise indicated

5. Collecting Food Samples

Food items suspected of causing foodborne illness can be tested for bacterial agents or toxins (but not viruses or most parasites). If a potential foodborne outbreak occurred, notify your local health department. CDPHE technical support may also occur via the Consumer Protection

Acceptable samples include: food consumed from the suspect meal; swabs of the food preparation area; food prepared in a similar manner as the suspect food; unopened samples of suspect canned or packaged food.

Obtain the materials outlined in 'General Materials for all Sample Collections' (page 16) and these items:

- Sterile plastic – food gloves
- Sample containers
- **Sterile plastic cups** with screw tops for liquids

When possible, use sterile utensils to collect the sample. If sterile utensils are not available, sanitize an appropriate utensil with an alcohol wipe or a sanitizer such as a 200-ppm bleach solution or a 400-ppm quaternary ammonia sanitizing solution.

An adequate sample quantity is approximately 200g (1/3 to 1/2 lb).

Collection Procedures – For Solid/Liquid Food

1) Place the food product in a sterile Ziploc® bag or Whirlpak®, or sterile container
   - Do NOT touch or handle the inside of the sample bag/container
2) Tightly seal the bag or container to prevent leakage
3) Ship food samples as follows:
   - Ship frozen foods on dry ice
   - Ship perishable or cold foods on ice or with refrigerated packs
   - Ship canned or low moisture foods at room temperature
2) Pack samples in an insulated box (***Packaging and Shipping*** section)
3) Specimens must be received at the laboratory within three days of collection

The CDPHE Lab will <u>not</u> accept sample containers with dimensions exceeding 11 ½ inches in size

6. Collecting Dead Bird Samples - West Nile Virus Testing

Birds provide early warning of West Nile virus (WNV) activity and can determine areas of increased risk of human exposure. Obtain a sample within 48 hours of the bird dying.

Obtain the materials outlined in 'General Materials for all Sample Collections' (page 16) and these items:

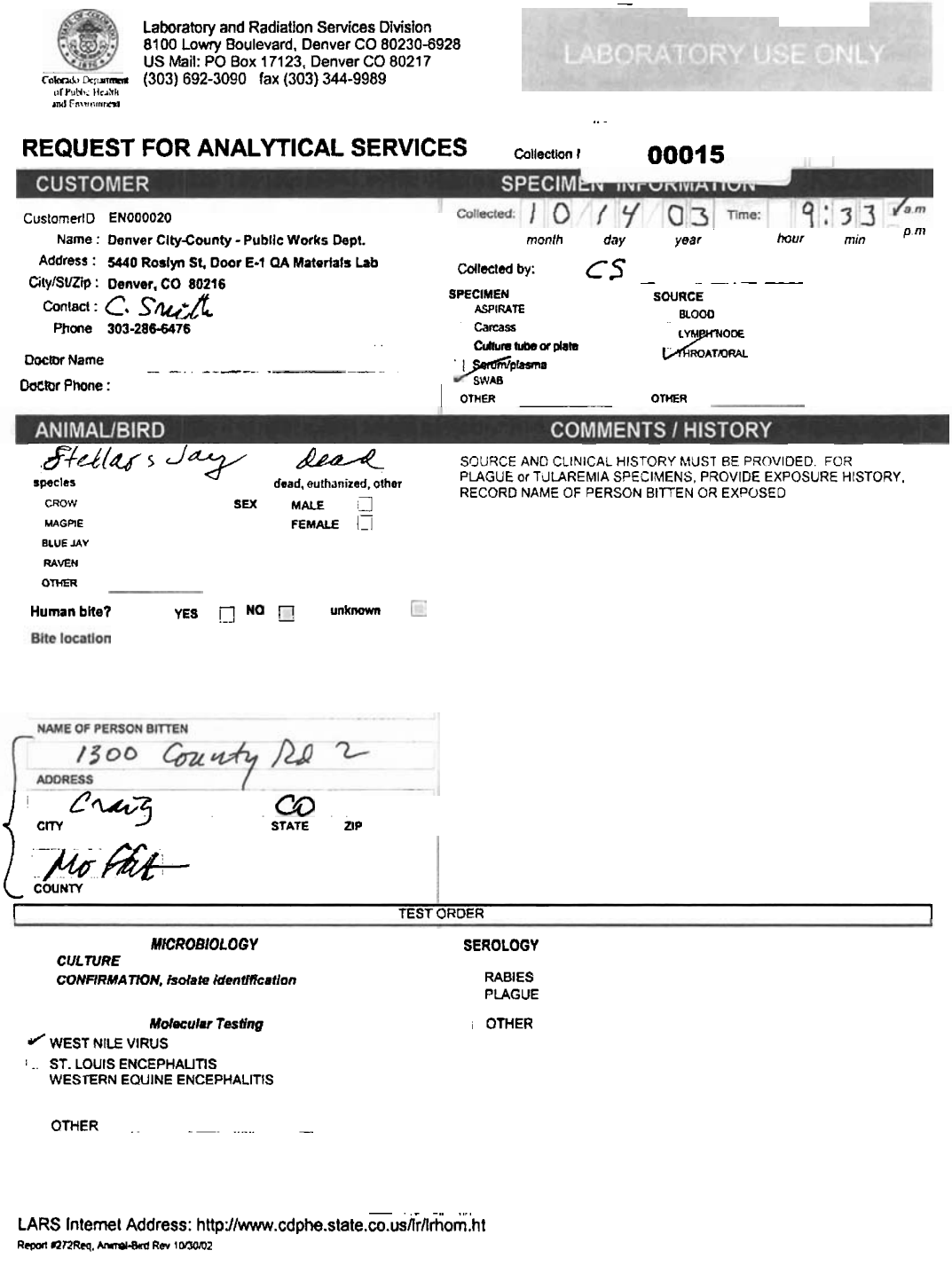
- Eye protection/goggles to protect from airborne material during swabbing
- Trash bag – for disposal of bird carcass
- Optional: Respirator (N95) – for covering the nose and mouth
  Note: The above materials are supplied by local public health for 'Avian Oral Swab Collection'
- 1 – Instruction Sheet
- 1 – Map of regional testing laboratories and county designations
- 20 – Dacron swabs
- 20 – Screw-cap tubes, 1.5 ml
- 10 – Shipping containers (double-mailer)
- 10 – Biohazard labels
- 10 – Diagnostic specimen labels
- 1 – Sheet Sample Collection Number labels
- 10 – Avian Swab Requisition Form 272 (*see **Figure1: Requisition Form 272***)
  Note: The above materials are supplied by CDPHE (303-692-3074) – 'Bird Kit' (10 pack)

Collection Procedures – For Dead Birds/West Nile Virus

1) Put on gloves and face/eye protection (respirator use is optional)
2) Open the bird's beak and check for any maggots or signs of decay
   - If maggots/decay is present do NOT obtain a sample
3) Swab the interior of the bird's beak and throat (under tongue as well) with a Dacron swab
4) Place the tip of the swab into the screw-cap tube
   - Break the shaft from the swab so the it fits in the vial
5) Seal the tube with the cap and tighten securely
6) Place the screw-cap tubes in the metal sample container
   - Add strips of newspaper/paper towel to prevent movement of tubes in the container
7) Put the metal sample container into the cardboard mailer
8) Complete Requisition Form 272 - **Samples with incomplete forms will not be tested**
   - Indicate the bird species and all other sample information requested
   - Classify the bird type using the identification chart; if no clear match, check 'other'
9) Transfer the sample collection number label to Requisition Form 272
   - Duplicate sample collection number label to each of the two 1.5 ml screw-cap sample collection tubes

Change gloves between sample collection to prevent cross-contamination and dispose of discarded gloves in an appropriate biohazard waste container.

Figure 1: Requisition Form 272 - For West Nile Virus Testing



Colorado Department of Public Health and Environment

Laboratory and Radiation Services Division
8100 Lowry Boulevard, Denver CO 80230-6928
US Mail: PO Box 17123, Denver CO 80217
(303) 692-3090 fax (303) 344-9989

LABORATORY USE ONLY

REQUEST FOR ANALYTICAL SERVICES Collection # **00015**

CUSTOMER

CustomerID EN000020
Name : Denver City-County - Public Works Dept.
Address : 5440 Roslyn St, Door E-1 QA Materials Lab
City/St/Zip : Denver, CO 80216
Contact : C. Smith
Phone 303-286-6476

Doctor Name
Doctor Phone :

SPECIMEN INFORMATION

Collected: 10 / 14 / 03 Time: 9:33 ✓a.m p.m
month day year hour min

Collected by: CS

SPECIMEN
ASPIRATE
Carcass
Culture tube or plate
✓ Serum/plasma
SWAB
OTHER

SOURCE
BLOOD
LYMPH NODE
✓ THROAT/ORAL
OTHER

ANIMAL/BIRD

Stellar's Jay — species
dead — dead, euthanized, other

CROW
MAGPIE
BLUE JAY
RAVEN
OTHER

SEX MALE ☐ FEMALE ☐

Human bite? YES ☐ NO ☐ unknown ☐
Bite location

COMMENTS / HISTORY

SOURCE AND CLINICAL HISTORY MUST BE PROVIDED. FOR PLAGUE or TULAREMIA SPECIMENS, PROVIDE EXPOSURE HISTORY. RECORD NAME OF PERSON BITTEN OR EXPOSED

NAME OF PERSON BITTEN

1300 County Rd 2
ADDRESS

Craig — CITY
CO — STATE
ZIP

Moffat
COUNTY

TEST ORDER

MICROBIOLOGY
CULTURE
CONFIRMATION, isolate identification

Molecular Testing
✓ WEST NILE VIRUS
ST. LOUIS ENCEPHALITIS
WESTERN EQUINE ENCEPHALITIS

OTHER

SEROLOGY

RABIES
PLAGUE

OTHER

LARS Internet Address: http://www.cdphe.state.co.us/lr/lrhom.ht
Report #272Req, Animal-Bird Rev 10/30/02

Chart 2: Instructions for Completing Requisition Form 272

All items listed below must be completed except for the Comments/History section.

Blank items on 272 Requisition Form	Information Required
Blank below Laboratory Use Only **Required**	Affix the sample collection number to both the sample collection tubes and Requisition Form #272
Customer; preprinted **Required**	Enter the contact name and phone number if blank
Specimen Information **Required**	Fill in the collection time fields indicating month, day, year and time Enter the name of the person collecting the specimen Enter the type of specimen Enter if sample is throat/oral for submission of bird swabs
Animal/Bird **Required**	Enter the identity of the bird species Note if the bird was live, euthanized, or dead when found Ignore the blank that is labeled "Name of Person Bitten" and enter the address, city, state, ZIP, and county blanks
Comments/History **Optional**	Enter additional information related to the sample or collection site that may be of epidemiological value (optional)
Test Order (left-hand side) **Required**	Enter 'West Nile Virus testing'

Ship the sample container via UPS or Greyhound bus service to the appropriate laboratory. Record the account numbers provided to the designated county point-of-contact.

Test results are mailed to the submitting agency and electronically sent to the county point-of-contact. Notification to the agency or individual who submitted the dead bird will be the responsibility of the local health department.

The CDPHE Lab will <u>not</u> accept items with dimension exceeding 11 ½ inches in size

7. Collecting Samples - Rabies Testing

Rabies confirmation analysis can be performed at CDPHE's laboratory and Colorado State University (CSU in Fort Collins) Diagnostic Laboratory. Technical consultation on rabies exposure, testing, bite follow-up and reporting, and rabies pre- and post- exposure prophylaxis. Can be obtained through the Disease Control and Environmental Epidemiology Division. Local public/environmental health agencies and animal control agencies can assist in preparing rabies samples.

CDPHE's Disease Control and Environmental Epidemiology Division must be notified of all samples to be tested for rabies: 303-692-2700; 303-370-9395 for nights/weekends.

Note: *Samples received by the CDPHE laboratory before 11:00 A.M. on a regular business day will be processed that same day. Those samples received after 11:00 A.M. will be processed the next workday, unless an emergency involving human exposure exists.*

Positive rabies results are telephoned immediately to the submitting agency and to the Disease Control and Environmental Epidemiology Division; negative results are reported by mail. Submitting agencies may contact the CDPHE Laboratory Services Division for results during regular business hours at 303-692-3485 or 303-692-3499. The CSU Diagnostic Lab can be reached at 970-491-1281.

Domestic Animal Samples

Appropriate domestic animal samples include dogs, cats, ferrets, livestock, etc. Submission of domestic animals to CDPHE is limited to those involved in human exposure. Dogs and cats that have bitten a person are required under Colorado law to be held for a 10-day observation period, as opposed to euthanizing and testing the animal. Any exception to this requirement must have prior approval from the CDPHE. If the biting animal remains healthy for the 10-day quarantine, the risk of rabies transmission is eliminated. Rabies testing is then unnecessary.

Wildlife Samples

Appropriate wild animal samples include carnivorous, terrestrial mammal species and bats involved in human or domestic pet exposure. With prior approval, bats, skunks, or wildlife exhibiting neurological symptoms are accepted for surveillance purposes. Rodents, rabbits and hares, wild or domestic, are not involved in rabies transmission, rarely ever infected, and are not accepted for rabies testing at CDPHE.

Appropriate samples acceptable for submission for rabies testing:

Bats: Submit the entire animal

- Dogs, cats, skunks, raccoons and similarly sized animals: Submit the head only
- Livestock: Submit the brain only

Collecting Procedure – Rabies Testing

1) Removal of heads or brains should be performed by individuals with knowledge and adequate personal protective equipment (PPE) to protect from rabies virus exposure
2) Place the sample in a plastic bag and seal; place the bag in a second bag and seal
   - Refrigerate sample immediately after collecting; hold at 35-40°F (2-8°C)
3) Complete a Rabies Epidemiology Form (available from local health departments, animal control agencies or CDPHE)
4) Label the bag containing the sample with the number on the Rabies Epidemiology Form
   - Attach a Rabies Epidemiology Form to each sample
5) Place sample in an inner waterproof container with cold packs
   - Do not use dry or wet ice
   - Do not freeze or place sample in a preservative (such as Formalin)
6) Place the container in an outer shipping container; mark as "Biological Specimen"

Note: It is the responsibility of the submitting agency to ensure no leakage during shipment.

Rabies samples may be shipped by bus, airfreight or overnight delivery to the CDPHE Laboratory. *DO NOT SEND BY MAIL* unless overnight delivery is guaranteed.

The CDPHE Lab will not accept items with dimensions exceeding 11 ½ inches in size

Chart 3: Quick Reference – Rabies Testing Submission

Rabies Submission		
Domestic animals	Horse or Bovine (head) - Have the vet remove the brain prior to submission	
Wildlife	Human Exposure - Emergency Status when skunks or bats involved - Call Epidemiology for surveillance	(303) 692-2628
Preparation	Use safety protection - Seek an expert (vet) for decapitation	
Shipment	Do not freeze – ice packs only Complete rabies form Courier or bus Prevent leakage of packing container	
Test results	Positive resulted are telephoned to submitter Negative results are sent by mail (can call)	(303) 692-2628

8. Collecting Samples - Plague Testing

Chart 4: Quick Reference – Plague Testing Submission

Plague Submission	
Rodents	Whole carcass, freeze if decay is evident. Double bag in clear zip-top bags; ship to CDPHE
Cats & dogs	Lymph node aspirate, or abscess aspirate, swab; if expired, liver and spleen tissue, in syringe with needle removed or sterile vial. Ship to CDPHE
Fleas	Place pool in plastic vial as per WNV mosquito pools, ship to CDPHE

9. Collecting Samples - Mosquito Trapping

Obtain the Mosquito Kit from CDPHE (303-692-3074)

Mosquito Kit (20-pack):

- 1 – Instruction sheet
- 20- 2 ml Screw-cap sample collection tubes (green top), pre-filled with ceramic beads
- 4 – Shipping containers (double-mailer)
- 4 – Return address labels – Molecular Science
- 4 – Biohazard labels
- 4 – Diagnostic specimen labels
- 1 – Sheet of Sample Collection Number labels
- 4 – Requisition Forms 273

Also obtain the following materials:

- Triethylamine (TEA) - optional
- 13 gallon plastic trash bag
- Collection nets
- Fan
- Cooler
- Dry ice
- Newspaper
- Clean white plastic tray

General Mosquito Collection

1. Gather traps early in the morning (to minimize damage/morbidity to mosquitoes)
2. Pinch off the collection net while the fan is still running
3. Record the following information on a piece of white medical tape:
   - Trap identification number, location/site
   - Sample collection date
   - Method of collection (i.e. light trap or gravid trap)
   - Collector's name
4. Place the tape on the capture net for identification and later reference

Mosquito Virus Testing

Mosquito testing is currently limited to *Culex* species through the CDPHE laboratory. Before mosquito identification and further testing, anesthetize the mosquitoes. Achieve this by cooling the samples in the collection net: place a small amount of dry ice inside a cooler insulated with newspaper (30-45 minutes) or freeze mosquitoes immediately by placing the collection nets directly on dry ice. *Note: If the nets have dew on them, freezing will damage mosquitoes caught in the frozen dew.* An alternative process involves using Triethylamine (TEA) can to permanently paralyze mosquitoes. This process avoids a freeze-thaw cycle that is deleterious to virus isolation. TEA is caustic, flammable and is very hazardous when inhaled. Thus, it is important to follow appropriate personal safety precautions.

After the mosquitoes are anesthetized or killed:

1. Transfer the mosquitoes from the collection nets to the sample tubes
2. Properly label each tube with the trap site information
3. Freeze the sample tube on dry ice
4. Empty the contents of collection net into a white plastic tray where mosquitoes are separated from non-mosquito captures and placed into the specimen vial
   - Anesthetized mosquitoes will stay down (about 10 minutes) for the transfer step
5. Identify and separate mosquitoes by species into pools (max. 50 mosquitoes per pool)
   - Perform this task as soon as possible to minimize damage to mosquitoes
6. Place sorted mosquitoes in the supplied screw-cap sample collection tubes
   - The sample collection tubes contain ceramic beads, which are a critical component of the processing of the samples at the laboratory. Carefully uncap the sample collection tube when placing the sorted mosquitoes into the tube. Securely re-fasten the cap.
7. Attach a Sample Collection ID number to the vials and to the Requisition Form
   - Use the page of Sample Collection ID number supplied in the Mosquito Kit (four copies of each number are provided)
8. Maintain a mosquito collection and identification record to record the following for all captured mosquitoes: collection/trap date; county; site identification; trap method
9. Complete the CDPHE Requisition Form 273 (*sample form on next page*)
   - Fill in the all fields on Requisition Form 273 (can record up to 5 pools per form)

Failure to complete the form may result in delays in testing or disposal of samples

10. Place the sample tubes and the completed Requisition Form 273 in the metal container
11. Put the metal container into the cardboard mailer and affix labels
    - Include return address, biohazard and diagnostic specimen labels
    - Ship via UPS or Greyhound bus service delivery to the Molecular Science laboratory

    Note: When assembling double mailers for shipment, be certain to insert the black rubber gasket into the lid of the inside canister.

Do not ship samples after Thursday or the day before a holiday to avoid having them delivered over the weekend or on a holiday; the lab will not set up samples until the next standard work day.

The CDPHE Lab will <u>not</u> accept items with dimensions exceeding 11 ½ inches in size

<u>Mosquito Samples - CDC Light Trap Method</u>

CDC Light Traps are used to capture female mosquitoes (species *Cx. Tarsalis*) seeking a blood meal. Gravid Traps are used to capture mosquitoes (species *Cx. pipiens* complex) that carry diseases impacting humans, including equine encephalitis and West Nile

In addition to the above items, obtain the following materials before collecting samples:

- Large padded manila envelope, newspaper, insulated plastic thermos jug w/ holes in bottom or insulated re-usable flexible nylon lunch bag w/ holes punched in the bottom

<u>Collection Procedure - CDC Light Trap</u>

1. Bait the trap with two to three pounds of dry ice per night of trapping
    - Actual quantities vary based on humidity and temperature
    - The dry ice can be placed in a large padded manila envelope (no holes), tightly wrapped in newspaper, in an insulated plastic thermos jug w/ holes drilled in bottom (keep spout open when using jug to keep condensate from freezing or plugging holes) or an insulated re-usable flexible nylon lunch bag w/ holes punched in bottom
2. Place the light trap 5 to 6 feet above the ground
3. Select a location for the light trap that is protected from competing light sources, smoke/ fume emitting areas (like industrial plants), high winds, public view or morning sun
    - Collect samples on non-full moon nights when using light traps
4. Place the light trap in an open area near good mosquito resting surfaces
    - Mosquitoes are located near abundant vegetation (i.e. trees, shrubs, sheds, stables, sewers/culverts, etc.) and /or areas where birds congregate (e.g. grain storage, livestock feeding areas, etc.)
5. Suspend the light trap below the padded envelope/newspaper holding the dry ice or from the bottom of the jug/lunch bag and then from a tree limb
6. Place a 13 gal plastic trash bag over the collection net (to protect mosquitoes from rain)
    - Cut a hole in the bottom of the bag to fit over the collection net

Consider collecting samples through this method for a minimum of two consecutive nights to maximize obtaining mosquitoes and minimize the influence of adverse weather.

Mosquito Samples – Gravid Trap Method

In addition to the above general sample collection materials, obtain the following materials for the Gravid Trap sample collection method:

- 38 liters unchlorinated (e.g. well water) water
- 1 pint undiluted Microbe Lift (commercial bacterial additive)
- 8 kg freshly cut bulrush
- 700 g dry horse manure
- Large plastic 15 gallon tub
- Breathable or vented container
- Permanent Marker
- Unchlorinated water

Mosquito Collection Procedure – Gravid Trap

1 **Bait trap with a fermented infusion of water** and manure (provided by Colorado Mosquito Control, Inc) using the following materials:
    - 38 liters unchlorinated (e.g., well water) water
    - 1 pint undiluted Microbe Lift (commercial bacterial additive)
    - 8 kg freshly cut bulrush
    - 700 g dry horse manure
2 Mix the bait ingredients thoroughly in a large plastic Rubbermaid 15-gallon tub
    - **Transfer the mixture to a breathable or vented container; mark the fill level** of the infusion on the container and let stand for 10 days
    - Periodically add unchlorinated water to marked fill level to adjust for evaporation
3 Fill the trap reservoir with the fermented mixture
    - **Fill to within 1 to 1.5 inches of the bottom** of the vertical suction tube
    - Consider drilling an overflow hole into the wall of the reservoir tub at the maximum infusion level (to keep the level of the infusion below the opening due to rain)
4 Add the mixture as necessary to maintain the space at the bottom of vertical suction tube
    - Change the mixture in the gravid trap when it begins to produce mosquito larvae
5 Place the Gravid trap in a location that is protected from the morning sun, near mosquito resting areas (e.g. abundant vegetation, outbuildings, sheds, sewers/culverts, etc.) and areas where birds congregate (e.g. grain storage, livestock feeding areas, etc.)
    - **Do not place trap near sites that compete (e.g. adjacent to a livestock water tank)**
6 Place a 13-gallon plastic trash bag over the collection net to protect mosquitoes from rain
    - **Place holes in bag to allow air to vent out**
7 Collect samples a minimum of two consecutive nights to maximize the sample

Figure 2: Request for Analysis of Mosquito Pool(s)

Colorado Department of Public Health and Environment

Laboratory and Radiation Services Division
8100 Lowry Boulevard, Denver CO 80230-6928
US Mail: PO Box 17123, Denver CO 80217
(303) 692-3090 fax (303) 344-9989

REQUEST FOR ANALYTICAL SERVICES

CUSTOMER

CustomerID PH002027
Name : Granby Veterinary Clinic
Address : PO Box 425
City/St/Zip : Granby, CO 80446
Contact :
Phone 970-887-3848
Contact Name : C Smith
Contact Phone : 970-887-3840

SPECIMEN INFORMATION

Collected by: CES
Comments:

MOSQUITO POOL DESCRIPTION

Accession # USE ONLY	LAB	Sample Collection #	Pool number	Collection date MM/DD/YY	Collection SiteID	County	method Lite vs gravid	genus/ species
		00016	LV-23	08-24-03	12	GRAND	lite	Culex tarsalis
		00017	LV-24	08-24-03	10	LAKE	lite	Culex tarsalis
		00018	LV-25	08-24-03	14	LAKE	lite	Culex pipiens
		00019	LV-26	08-25-03	16	GILPIN	gravid	Culex tarsalis
		00020	LV-27	08-25-03	8	GRAND	lite	Culex tarsalis

SAMPLE
DO NOT USE

LARS Internet Address: http://www.cdphe.state.co.us/lr/lrhom.ht
Report #273 MOSQUITO POOL Req. 10/30/02

PACKAGING AND SHIPPING SAMPLES

Background

The United Nations, Federal agencies, United States Postal Service (USPS,) and private carriers strictly regulate the packaging and shipment of biological, chemical and radiological specimens to ensure the safety of their workers, the public and the package recipients. Before sending specimens to the CDPHE LSD for testing, submitters must determine the following:

- Type of specimen (powder, blood, tissue, chemical, radioactive, etc)
- Type of analyses to be performed (biological, chemical, radiological)
- Quantity of the specimen needed for testing
- Classification of the specimen as a hazardous/infectious material.
  - The CDPHE Laboratory must be notified prior to shipping such samples
- Temperature required to preserve the specimen during transit
- Specimen packaging size and containment; CDPHE standards are met
- Time frame for specimen arrival at the CDPHE laboratory
- Whether the specimen is evidence in a criminal investigation
- Chain of custody documentation

The CDPHE laboratory does <u>not</u> accept items larger than 11 ½ inches in size

Contact the CDPHE Laboratory Services Division at 303-692-3090 for help with information about the appropriate packaging and shipping of a specimen.

A. Packaging Samples For Shipping

1. Non-Infectious Samples - U. S. Postal Service Mailing

This section pertains to Infectious or Non-Infectious samples mailed to laboratories.

a. <u>Mailing Samples in Tubes</u>

Primary Container:

Note: The specimen tube is the primary container.

- Write the specimen ID number on the side of the specimen tube
- Cover the specimen tube with cellophane tape
- Use a strip of Para film to wrap and seal the lid/cap interface
- Wrap the specimen tube in an absorbent material (paper towels)

Secondary and Tertiary Container:

Note: The inner tube is the secondary container; outer tube is the tertiary container.

- Insert the wrapped sample tube into the inner/secondary mailing tube
- Cap the secondary tube
- Fold up paperwork and wrap it around the inner tube
- Place the secondary tube and paperwork inside the outer/tertiary mailing tube

(1) Labeling Shipping Tube Containers

- Attach a *To/From* label to the outside of the container.
- On the label, include the name and address of the shipper and receiver **plus** the name and the telephone number of the person responsible for the sample
  - CDPHE prefers the responsible party records a 24-hr contact number
- Print 'Micro 0622' (a CDPHE mail code) under-the return address
- Affix at least one red *BIOHAZARD* label to the outer shipping container
- If the sample is a known organism, record the organism name or the word 'Bacteria' on the red *BIOHAZARD* label
- If the sample is a medical or diagnostic sample, write 'Medical,' 'Clinical,' or 'Diagnostic Specimen' on the *BIOHAZARD* label

b. Mailing a Sample Plates

Do not use a standard double mailer to send a specimen plate.

Primary Container:
Note: The specimen plate is the primary container.

- Write sample number on the face of the plate and cover with cellophane tape
- Use a strip of Para film to wrap and seal the lid/cap interface
- Wrap the plate with an absorbent material, such as paper towels
- Use packing material to secure the inner contents from movement

Secondary and Tertiary Containers:
Note: The inner container is the secondary container. This is typically a cardboard box small enough to fit into the tertiary container, also a cardboard box. It may also be a specially made "plate mailer" usually made of Styrofoam.

- **Insert the wrapped plate into the secondary container and seal tight**
  - Use packing material to secure the inner contents from movement
- Fold paperwork and attach it to the inner container
- Place the secondary container and paperwork inside the tertiary container

(1) Labeling Shipping Containers

- Attach a *To/From* label to the outside of the container
- On the label, include the name and address of the shipper and receiver **plus** the name and telephone number of the person responsible for the sample.
  - CDPHE prefers the responsible party records a 24-hr contact number
- Print 'Micro 0622' (CDPHE mail code) under-the return address.
- Affix at least one red BIOHAZARD label to the outer box.
- If the sample is a known organism, record the organism name or the word 'Bacteria' on the red *BIOHAZARD* label
- If the sample is a medical or diagnostic sample, write 'Medical,' 'Clinical,' or 'Diagnostic Specimen' on the *BIOHAZARD* label

2. Non-Infectious Samples - Overnight Mailing

This section pertains to overnight mailing via FedEx shipments. If other couriers are used, contact courier for overnight mailing guidelines for samples.

a. Overnight Mailing Non-Infectious Substances - Tubes

Primary Container:

Note: The sample tube is the primary container.

- **Write sample number on the side of the sample tube**
- Cover with cellophane tape
- **Use a strip of Para film to wrap and seal the lid/cap interface**
- Wrap the specimen tube in absorbent material (paper towels)

Secondary and Tertiary Containers:

Note: The inner tube (usually made of metal) is the secondary container; the outer mailing container is the tertiary container.

- **Insert the wrapped sample tube into the secondary container; cap the mailing tube**
- Fold paperwork around the container
- Place the secondary container and paperwork inside the outer mailing tube
- Over pack secondary container inside a standard FedEx box (tertiary container)
  Note: See Over pack instructions below.
  - The outer box must be large enough to accommodate the FedEx Air Waybill pouch without folding it
  - The Air Waybill must be visible to read without removing it from the pouch; it must be easy to remove and insert

(1) Labeling Overnight Shipping - Tubes

- Attach a *To/From* label to the outside of the container
- On the label, include the name and address of the shipper and receiver **plus** the name and telephone number of the person responsible for the sample
  - CDPHE prefers the responsible party records a 24-hr contact number
- Print 'Micro 0622' (CDPHE mail code) under the return address on label
- Attach FedEx AIR WAYBILL pouch to the outside of the double mailer -
  - Make sure the Air Waybill will fit, without folding, and is visible to read without removing it from the pouch
  Note: See the section below on completing the Air Waybill.
- For medical, clinical or diagnostic samples, use a standard FedEx air waybill The Dangerous Goods Air Waybill is not necessary
  - Once complete, insert the Air Waybill into the pouch
- Affix at least one red *BIOHAZARD* label to the outer tube
- Write 'Medical,' 'Clinical,' or 'Diagnostic Specimen' on the *BIOHAZARD* label for a medical or diagnostic specimen

b. Overnight Mailing Non-Infectious Substances – Sample Plates

Do not use a standard double mailer to send a specimen plate.

Primary Container:
Note: The plate is the primary container.

- **Write the sample number on the face** of the plate and cover with cellophane tape
- Wrap the plate with an absorbent material, such as paper towels
- Use packing material to secure the inner contents from movement
- Use a strip of Para film to wrap and seal the lid/cap interface

Secondary and Tertiary Container:
Note: The inner container is the secondary container (typically a cardboard box or a 'plate mailer' – usually Styrofoam – that is small enough to fit into a larger box.

- **Insert the wrapped plate into the secondary container and seal**
- Fold paperwork and attach it to the secondary container
- Place the secondary container and paperwork inside the another container; seal
  - If necessary, use packing material between the two boxes
- Pack this container inside a FedEx box (tertiary container)

(1) Labeling Overnight Shipping – Sample Plates

- Attach a *To/From* label to the outside of the container. On the label, include the name and address of the shipper and receiver **plus** the name and telephone number of the person responsible for the sample
- Print Micro 0622 (CDPHE mail code) under the return address
  Attach FedEx AIR WAYBILL pouch to the outside of the double mailer
  - Make sure the Air Waybill will fit, without folding, and is visible to read without removing it from the pouch
    Note: See the section below on completing the Air Waybill.
- See the section below on completing the Air Waybill; insert it into the pouch
- Affix at least one red *BIOHAZARD* label to the outer box.
- Write 'Medical,' 'Clinical,' or 'Diagnostic Specimen' on the *BIOHAZARD* label for a medical or diagnostic specimen

3. Infectious Samples - U. S. Postal Service Mailing

This section pertains to mailing samples that contains an infectious material.

a. Mailing Samples

Primary Container:
If the sample contains a known or suspect etiologic agent, the outer or final packaging must display additional information and meet specific requirements.

No more than 50 ml/50 g of an infectious substance can be shipped in a single United Nations approved package or shipment. **Only United Nations approved packaging can be used for overnight shipments of infectious substances.**

Note: The fiberboard box in which the double or plate mailer is placed must be a tested and U.N. – approved system for containment of infectious substances.

- The box must exhibit U.N. performance markings that verify it meets the requirements (for example, "U.N. Class 6.2/95")
- Pack samples as detailed above for non-infectious agents
- There are no specific requirements for packing the mailer inside the U.N. box if the contents are secure and immobile

(1) Labeling Shipping

- Label as above, but do <u>not</u> cover up any U.N. markings
- Place an Air Waybill window pouch on the box in a manner that allows the Air Waybill to be easily removed and reinserted
  - The Air Waybill must be readable through the window and not folded
- The *To/From* label should contain the same information as regular mailings
- The *To/From* label and the FedEx Air Waybill must be the same
- On the outside edge of the "Infectious Substance" diamond (the design stamped on the box), hand print:
  'Affecting Humans; Bacteria: (<u>name of predominant bacteria</u>)" **and**
  'U.N. 2814'
- Affix at least one red *BIOHAZARD* label to the outer tube or box
  - If the sample is a known organism, write in its name (bacteria) or the word "Bacteria" on the red label

Special U.N. – approved boxes state "Infectious Substance" on them and the performance markings. However, the information mentioned above still must be added as described.

All FedEx shipments must be in packaging that will accommodate the Air Waybill and pouch so that the Air Waybill will fit in the pouch without folding, can be easily inserted or removed, and can be read without being removed.

3. Completing the Air Waybill

a. <u>Non-Infectious Substances</u>

If the sample is a medical or diagnostic specimen, use the standard Air Waybill supplied by FedEx.

b. <u>Infectious Substances</u>

If the sample is an infectious substance, use the "Dangerous Goods" Air Waybill.

- Account number and date must be on the Air Waybill
- Fill out the "*To*" and "*From*" boxes completely, including phone numbers
  - The two boxes must match the *To/From* label on the package
- For normal overnight delivery, check the "FedEx Standard Overnight" box under Section 4a
  - Do <u>not</u> FedEx on a Friday unless the business is open on Saturday to receive it

- Check '*Dangerous Goods as per attached Shipper's Declaration*' box under Section 6
- If the account number listed in section 1 is to be billed, check the "*Sender*" box in Section 7
- Fill in "*Total Package*s" and "*Total Weight*" blanks. Use estimated weight in the Total Weight blank. For shipping purposes, consider each culture or specimen to contain approximately two (2) mg; two (2) tubes = four (4) mg, and so forth.
- Under "*Transport Details*," mark through the boxes stating "*Cargo Aircraft Only*" and "*Radioactive*"

Note: The bottom area of the Air Waybill must be accurate, or the courier will not deliver the package. If the headings below are not on your copy of the Air Waybill, print them in as shown on the example. For bacterial isolates or cultures, fill in the blanks as follows:

- Below "*Proper Shipping Name*," print: **Infectious Substance Affecting Humans**
- Under this, print "Bacteria: _____"
  - Fill in the blank with the name of the bacteria being sent.
  - If more than one isolate is sent, give the name of the predominant organism
- Below "*Class or Division*," print: **6.2**
- Below "*U.N*." or "*ID No*." print: **U.N. 2814**
- Below "*Quantity and type of packing*," print: **One Fiberboard Box X _____mg**
  - Fill in the blank with the weight of the specimen(s)
  - For shipping purposes, consider a sample to be about 2 mg (2 tubes = 4 mg, etc)
  - One shipment of infectious substance must <u>not</u> exceed 50 g or 50 mL
- Below "*Packing Inst*.," print: **602**
- Below "*Additional Handling Information*," print: **Prior arrangements as required by the IATA Dangerous Goods Regulations 1.3.3.1 have been made** (IATA Statement)
- Following the IATA statement, print the name and phone number of the person with primary responsibility for the sample
- Fill out the balance of the sheet as instructed
- The "*Place and date*" blank refers to name and location of receiving facility (Example: CDPHE; Denver, CO, + date.)
- In the "*NATURE AND QUANTITY OF DANGEROUS GOODS*" section, print: **Emergency Phone Number**
  - A 24-hr number for a person/business liable for any required emergency response regarding the shipment. (Example: Dr. smith 303-xxx-xxxx)

Figure 3: Diamond Printed On A FedEx Box

Figure 1 : FedEx Dangerous Goods Air Waybill

SHIPPER'S DECLARATION FOR DANGEROUS GOODS (Provide at least three copies to FedEx Express

Shipper	Air Waybill No Page of Pages Shipper's Reference Number
Consignee	FedEx Federal Express
Two completed and signed copies of this Declaration must be handed to the operator.	WARNING Failure to comply in all respects with the applicable Dangerous Goods Regulations may be in breach of the applicable law, subject to legal penalties. This Declaration must not, in any circumstances, be completed and/or signed by a consolidator, a forwarder or an IATA cargo agent.
TRANSPORT DETAILS This shipment is within the limitations prescribed for *(delete non-applicable)* PASSENGER AND CARGO AIRCRAFT / CARGO AIRCRAFT ONLY Airport of Departure	
Airport of Destination	Shipment type: *(delete non-applicable)* NON-RADIOACTIVE / RADIOACTIVE

NATURE AND QUANTITY OF DANGEROUS GOODS

Proper shipping name, Class or Division, UN Number or Identification Number, Packing Group (if required), and all other required information.

Proper Shipping Name	Class or Division	UN or ID No.	Packing Group	Subsidiary Risk	Quantity and Type of Packing	Packing Inst.	Authorization

Additional Handling Information

I hereby declare that the contents of this consignment are fully and accurately described above by the proper shipping name, and are classified, packaged, marked, and labelled/placarded, and are in all respects in proper condition for transport according to applicable international and national governmental regulations.	Name/Title of Signatory Place and Date Signature *(see warning above)*

IF ACCEPTABLE FOR PASSENGER AIRCRAFT, THIS SHIPMENT CONTAINS RADIOACTIVE MATERIAL INTENDED FOR USE IN, OR INCIDENT TO, RESEARCH, MEDICAL DIAGNOSIS, OR TREATMENT

4. Overpacking for Shipping

If the U.N. approved packaging is to be overpacked (repacked into a larger, non-specific box), state "**Overpack Used**" below the IATA statement at the bottom.

Overpacking can be done to provide a larger outer container to ensure the FedEx and From/To labels fit on the box. However, except for the container testing ~~r~~requirements the labeling requirements pertaining to Infectious Substances must be placed on the overpack box AND on the inner packaging.

To be safe when overpacking, print a statement on the outside confirming use of "U.N.-approved" packaging on the inside.

5. Other Packaging Systems

Complete 'Infectious Substance' packaging systems are available for purchase that are specially designed to meet all the shipping requirements. They include secondary containers (heavy plastic canisters) along with all necessary instructions and packing materials.

B. Other Shipping Methods

Private contract courier can be used to transport specimens to the CDPHE laboratory. These samples should pose no significant threat to the courier. Contact CDPHE laboratory for instructions.

Shipping rabies and other carcass samples may be shipped by bus. Contact CDPHE laboratory for instructions.

Appendix 1: Guidance on Initial Responses to a Suspicious Letter/Container With a Potential Biological Threat

This is an FBI – DHS – HHS/CDC Coordinated Document

A large number of potentially suspicious letters and packages continue to be reported to federal, state, and local law enforcement and emergency response agencies nationwide. In some instances these letters or packages may include powders, liquids, or other materials. Federal, state, and local response agencies should be mindful of the potential for small-scale exposure, which could result from material contained in threatening or suspicious packages. While this guidance is generally focused on the initial response to potential biological threats, all personnel responding to such incidents must be aware of the potential for exposure to hazardous chemical and/or radiological materials in addition to biological hazards. Additionally, there may be a threat posed from secondary releases or devices. Consistent with established protocols, response agencies should follow standard law enforcement procedures and hazard risk assessments in response to calls, and should pre-identify the relevant local public health points of contact to be notified in the event of a potential bioterrorism event.

The following guidelines are recommendations for local responders, based on existing procedures (including recommendations from the International Association of Fire Chiefs). This document provides guidance on the initial response to a suspicious letter/container, while other follow-on response plans, such as portions of the National Response Plan (NRP), may be utilized if a threat is deemed credible. In general, these potential threats or incidents fall into one of five general scenarios. They are as follows:

1. Letter/container with unknown powder-like substance and threatening communication (with or without illness):

Since there is an articulated threat, it is likely that the substance was intentionally introduced into the package in an effort to validate that threat. An articulated threat itself (with or without the presence of a suspicious substance) is a federal crime and may also constitute a violation under state and local statutes. The local Federal Bureau of Investigation (FBI) Weapons of Mass Destruction (WMD) Coordinator and/or FBI Joint Terrorism Task Force (JTTF), a certified HAZMAT unit, local law enforcement, and the local public health department should be notified. The role of Incident Commander (IC) will be assumed by the appropriate authority, as designated by state or local law. In many cases, the IC will be the most senior public safety officer (most likely the fire department chief or deputy chief, however, in many circumstances it may be a local sheriff or senior local or state police official). As such, it is the responsibility of the IC to establish the Incident Command System (ICS) and to ensure that notifications of the above-mentioned responders have been made or are in the process of being made. As the referenced agencies arrive, the IC will evolve into a Unified Command, as necessary.

At this stage, and later again as necessary, the FBI will conduct a timely WMD threat assessment with local law enforcement/fire/HAZMAT personnel. Depending on the nature of the threat, this assessment may include relevant interagency partners. This process utilizes coordination from FBI Headquarters elements to conduct an initial assessment of the credibility of the threat and provide technical support to responders who are on-scene. In coordination with recommendations from the threat assessment process and the unified command on-scene, an appropriately trained HAZMAT unit should screen evidence for the presence of chemicals and radiological material and double-bag in clear sealed bags (where possible), consistent with chain-of-custody requirements. Before packaging and when possible, photographs of the letter/container should be taken and relevant information should be documented, in coordination with the FBI WMD Coordinator. Under NO CIRCUMSTANCES should an unprotected responder, such as a law enforcement officer, attempt to package an unknown substance.

If this incident involves an unopened container such as a box, it must be evaluated by a certified bomb technician/explosives ordinance disposal personnel prior to being handled by HAZMAT. Any such letters/packages must also be evaluated by the HAZMAT unit for only a broad class of radiological and chemical threats prior to being released to law enforcement personnel for transport. This is required by the laboratory in an effort to protect the staff members who will ultimately be opening the container and performing definitive biological testing and/or forensic examinations.

The FBI, or the responding law enforcement agency, will ensure that a certified HAZMAT team has performed necessary field safety screening before transporting to an appropriate laboratory. This field safety screening should be clearly documented and limited to screening for pH (for liquids), radioactivity, volatile organic compounds, flammable materials, and oxidizing agents. Definitive analysis will only be performed by the appropriate laboratory.

A chain-of-custody form must be initiated along with an incident report. The FBI will then coordinate delivery of the evidence to the designated Laboratory Response Network (LRN) laboratory for further testing and analysis.

If individuals immediately present with illness in this scenario, the public health departments will have an increased role in the initial response. These issues are further addressed in the 'Critical Response Issues for Scenario #1' included below.

If the FBI Headquarters-led threat credibility assessment process deems the threat to be credible, the FBI will immediately notify the Centers for Disease Control and Prevention (CDC), the Department of Homeland Security Operations Center (HSOC), and other appropriate federal agencies. Appropriate response guidelines to a credible threat will be utilized from the NRP, including the Biological Annex and Terrorism Incident Law Enforcement and Investigation Annex. Depending on the nature and scale of the incident, the Department of Homeland Security (DHS) may choose to help coordinate response activities based on NRP procedures which, at a minimum, may include coordinating a joint public affairs statement.

2. Letter/container with a threat but no visible powder or substances present:

Merely threatening the use of a chemical or biological agent *is* a violation of federal law and merits investigation. As in scenario #1, all of the responders should be notified. Although no powder may be visible to the eye, there could be trace amounts of material present that could represent a health risk and also provide critical forensic evidence required for further investigation and prosecution. Therefore, the guidance in Scenario #1 also applies to responses to a letter/container containing a threat with no visible powder or substance.

3. Letter/container with unknown powder, no articulated threat, and no illness:

As there is no threat and no one is ill, it must be determined if there is a logical explanation for the presence of this substance. For example, HAZMAT teams have responded to a number of letters that contained crushed samples from vitamin and pain-relief companies. If a reasonable and defendable explanation can be given as to the source of the substance, that there is no articulated threat, and that no one is ill, then no further actions are necessary.

If, however, a reasonable source cannot be determined or there is any uncertainty, the steps outlined in scenario #1 must be conducted.

4. Letter/container with no visible powder, no threat, but recipients are ill:

This scenario has the most potential for ambiguity and confusion. Those who come in contact with *Bacillus anthracis* (anthrax), or other biological pathogens/toxins, may not immediately appear symptomatic. Although no powder or substance may be available to be collected for environmental testing, public health officials may decide to utilize clinical samples from potentially exposed individuals. Additionally, in this scenario it may be difficult to determine if a letter/container is actually associated with the illness. As there is no specific threat to investigate, this is primarily a public health and medical issue; but this scenario also represents a potential criminal act that should be jointly investigated by public health and law enforcement. The initial notifications will largely be the same as scenario #1, with public health taking a primary role in the response. While the primary concern is the treatment and well-being of the recipient, public health and law enforcement should maintain close contact, while public health determines the nature of the illness and law enforcement examines any relevant intelligence. Depending on the scale and nature of the incident, if HHS/CDC is notified they will maintain close contact and coordinate with DHS. If a potential criminal nexus is identified, the FBI will conduct an initial threat assessment and initiate appropriate actions and notifications listed under scenario #1.

5. Letter/container arrives with no powder, no threat, the recipient is not ill, but the recipient is concerned about the package:

With strict regard to federal criminal statutes, no investigative actions are necessary in this matter. However, if other threat indicators are present such as excess postage, misspelled names, unusual odors/colors, etc., law enforcement and the United States Postal Inspection Service should be notified to evaluate it for potential hazards. If the assessment determines that the letter/container is “suspicious,” then appropriate steps outlined in scenario #1 would be initiated.

Critical Response Issues for Scenario #1:

1. Request the assistance of the nearest certified hazardous materials response team to conduct risk assessments, field safety screening, sample (evidence) collection, decontamination, and other mitigation activities. Any sample (evidence) collection must be coordinated with law enforcement (FBI).

2. Notify appropriate law enforcement (local, state and local FBI WMD coordinator/JTTF, postal inspectors) when a potential threat is identified.

3. Do not touch, move, or open any suspicious package until an initial hazard risk assessment of the package can be performed in coordination with HAZMAT personnel and law enforcement.

4. An initial threat credibility assessment will be coordinated via the local FBI WMD Coordinator and the FBI Counterterrorism Division's Weapons of Mass Destruction Operations Unit (WMDOU). This will include the FBI Laboratory Division, Hazardous Materials Response Unit (HMRU) and other select interagency subject matter experts, tailored for the specific threat. This assessment includes an analysis of technical feasibility, operational practicability, behavioral resolve, and examination of any intelligence that might relate to the threat. If the threat is determined to be credible, other appropriate federal agencies will be notified, to include DHS and HHS/CDC. Additional information on this process is available from the NRP, including the Biological Annex and Terrorism Incident Law Enforcement and Investigation Annex.

5. Contact your local public health department (who should in turn notify state authorities and the CDC) if there is a threat of public health exposure or environmental contamination exists. HHS/CDC will then notify the HSOC, where appropriate.

6. In coordination with law enforcement, always notify the U.S. Postal Inspection Service, whenever it appears that the threat was delivered through the U.S. Postal Service. Assist with ensuring that origin and tracking information is obtained from the package (ideally, photographs of the front and back).

7. Treat the scene as a crime scene. Preserve evidence in coordination with law enforcement and ensure that materials are safely packaged. Take steps to retain enough suspicious material for:

   a. Laboratory analysis;
   b. Forensic examination of criminal evidence, regardless of whether the threat is ultimately determined to be accompanied by a hazardous material.

8. Transfer custody of evidence to a law enforcement officer as soon as possible. Maintain chain of custody by obtaining a record of names and signatures every time custody of a suspicious material or sample for laboratory analysis changes hands.

9. Perform basic field safety screening of the substance to rule out explosives, radiation, flammability, corrosives, and volatile organic compounds prior to transporting the materials to the appropriate LRN, as coordinated with the FBI WMD Coordinator. All field safety screening that is performed by responders should be clearly documented and shared with law enforcement and the LRN.

10. In coordination with the local FBI WMD Coordinator (and/or a responding law enforcement entity), transport samples to the designated CDC-qualified LRN facility. If field

safety screening detects the presence of chemical or radiological hazards, the FBI WMD Coordinator will contact FBI Headquarters for guidance regarding which laboratory is appropriate to perform the analysis. This will be done as part of the threat credibility assessment process noted above (see #4).

11. In coordination with public health and law enforcement, identify and list the names and contact information for anyone who may have been exposed to the suspicious substance so that they may be contacted when the LRN test results are available or if there is other additional information. If positive results are obtained, state and local public health departments will need to contact those potentially exposed as soon as possible to provide appropriate assistance (e.g., antibiotics, education, additional testing, vaccination, surveillance/symptom reporting).

12. In coordination with the FBI, identify a single point-of-contact for incident follow-up.

13. If LRN tests identify positive results for threat agents or a threat is determined to be credible, the FBI will immediately notify the DHS and other appropriate federal agencies to initiate relevant NRP actions, as necessary. The DHS will work closely with the FBI, HHS/CDC and other agencies to ensure a coordinated response.

Note on field screening

Once activities are complete to address immediate public safety concerns, every effort must be made to preserve evidence necessary for public health and law enforcement investigations.

> In situations where biological threat agents are suspected, the item(s) should be field safety screened and immediately transported in law enforcement custody to an LRN laboratory. This should be done in coordination with the local FBI WMD Coordinator.

Field safety screening should be limited to ruling out explosive devices, radiological materials, corrosive materials and volatile organic compounds. Currently, there are no definitive field tests for identifying biological agents. Additional field testing can mislead response efforts by providing incorrect or incomplete results, and destroy limited materials critical for definitive laboratory testing required to facilitate any appropriate public health and law enforcement response.

This information is provided for guidance. Questions related to the content of this document can be addressed to: Scott Steele, Ph.D., Counterterrorism Division,
WMD Countermeasures Unit, Federal Bureau of Investigation,
E-mail: ssteele2@leo.gov

Appendix 2: Hazardous Specimen Assessment Checklist

For credible or unknown threat specimens, first contact the FBI or local law enforcement agency for a threat assessment. Specimens must be pre-screened for radioactivity, volatile organic chemicals, explosives and other hazards prior to submission to the CDPHE Laboratory for further analysis. The CDPHE Laboratory must be notified before specimens are brought to the facility. The maximum allowable dimension of specimen packages that can be received by the CDPHE Laboratory is 11½ inches.

Submitter:

Name: **Title:** **Organization:**

Address:

Phone: **Fax:** **E-mail:**

Specimen:

Type:

Weight (g) or volume (ml):
Sample collection location:
Collection method:

Radioactivity assessment:

Is radiation present? Yes No
If yes, reading and unit of measurement:
Type of radiation detected:

pH assessment:

Testing device used: pH paper pH meter
pH level:

Volatile Organic Chemicals Assessment:

Is this type of chemical present? Yes No
If yes, type of chemical and concentration:

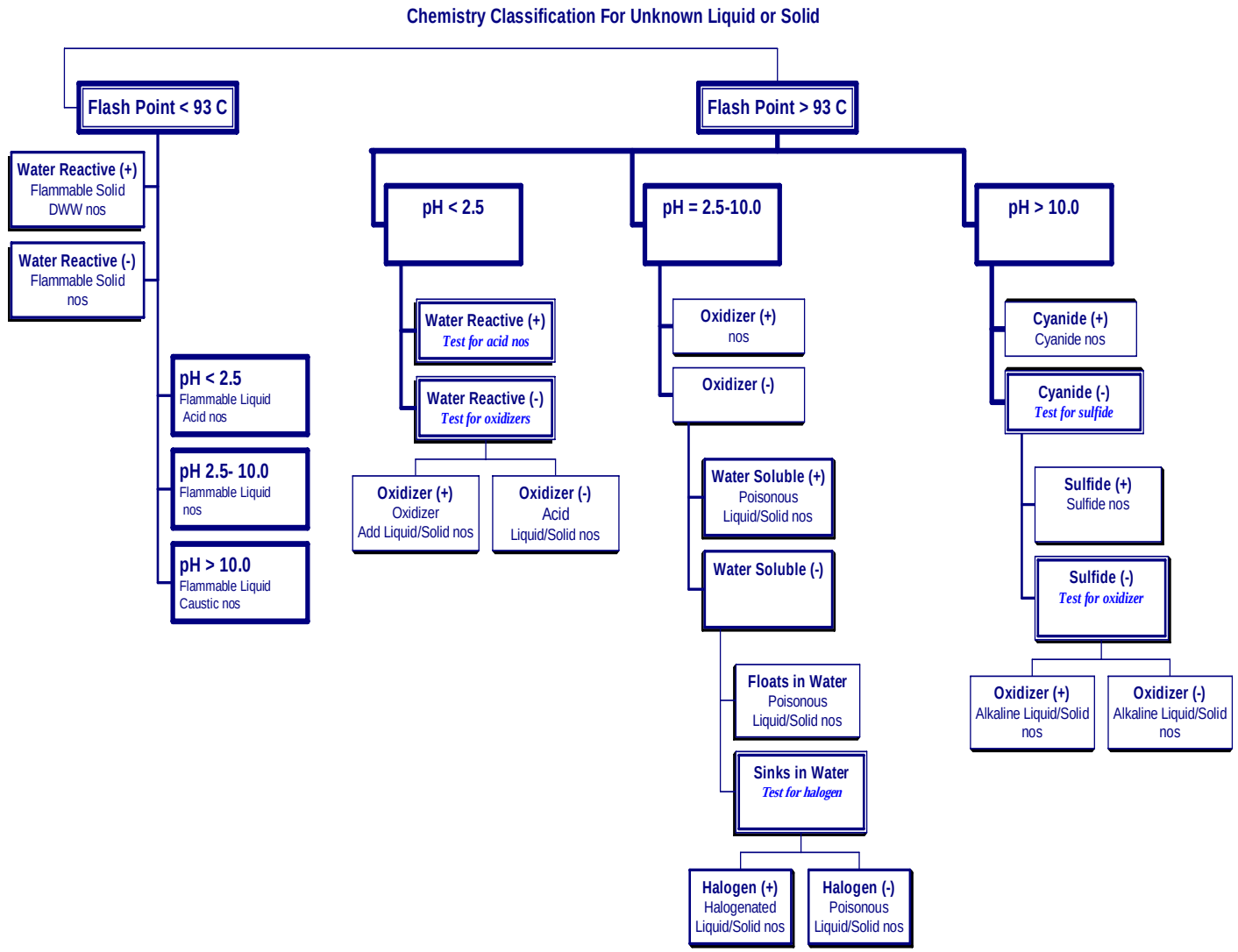
Incendiary/Aerosol/Dissemination Device Assessment:

Type of inspection conducted:	Visual		X-ray
Incendiary device detected?	Yes	No	
Aerosol device detected?	Yes	No	
Pressurized vessel present?	Yes	No	
Other dissemination device?	Yes	No	If yes, type of device:

This environmental specimen/package (ID#_____) has been assessed by individuals specifically trained or certified to perform the listed assessments. The assessments have been performed using acceptably calibrated/certified instruments or other acceptable means (as stated). Interpretation of these assessments indicates that the specimen/package has been declared free of hazardous levels of the following: radioactivity, volatile organic chemicals, incendiary or aerosol devices, pressurized vessels and other potential dissemination devices.

_____ ___/___/___ _____
Submitter Signature Date Time

Appendix 3: Chemistry Classification



Technical support is available both through the CDPHE Hazardous Materials and Waste Management Division and the Laboratory Services Division.

Appendix 4: Biological Testing Offered

CDPHE Laboratory Testing Availability for Biological Agents

Disease (Agent)	DFA*	PCR**	Culture	Toxin***	Antigen****	Serology
Anthrax (*Bacillus anthracis*)	X	X	X		X	
Botulism (*Botulinum* toxin)			X	X		
Brucellosis (*Brucella* spp.)		X	X		X	X
Cholera *(Vibrio cholerae)*			X			
Glanders (*Burkholderia mallei)*		X	X			
Melioidosis *(Burkholderia pseuodomallei)*		X	X			
Plague (*Yersinia pestis*)	X	X	X		X	
Ricin					X	
Tularemia (*Franciscella tularensis*)	X		X		X	X
SEB (Staph enterotoxin B)					X	
Q fever (*Coxiella burnetii*)					X	X
Rash illness testing						
Orthopox		X				
Vaccina (vaccine virus)		X				
VARIOLA (SMALLPOX VIRUS)		X				
Varicella Zoster (Chickenpox & Shingles virus)		X				

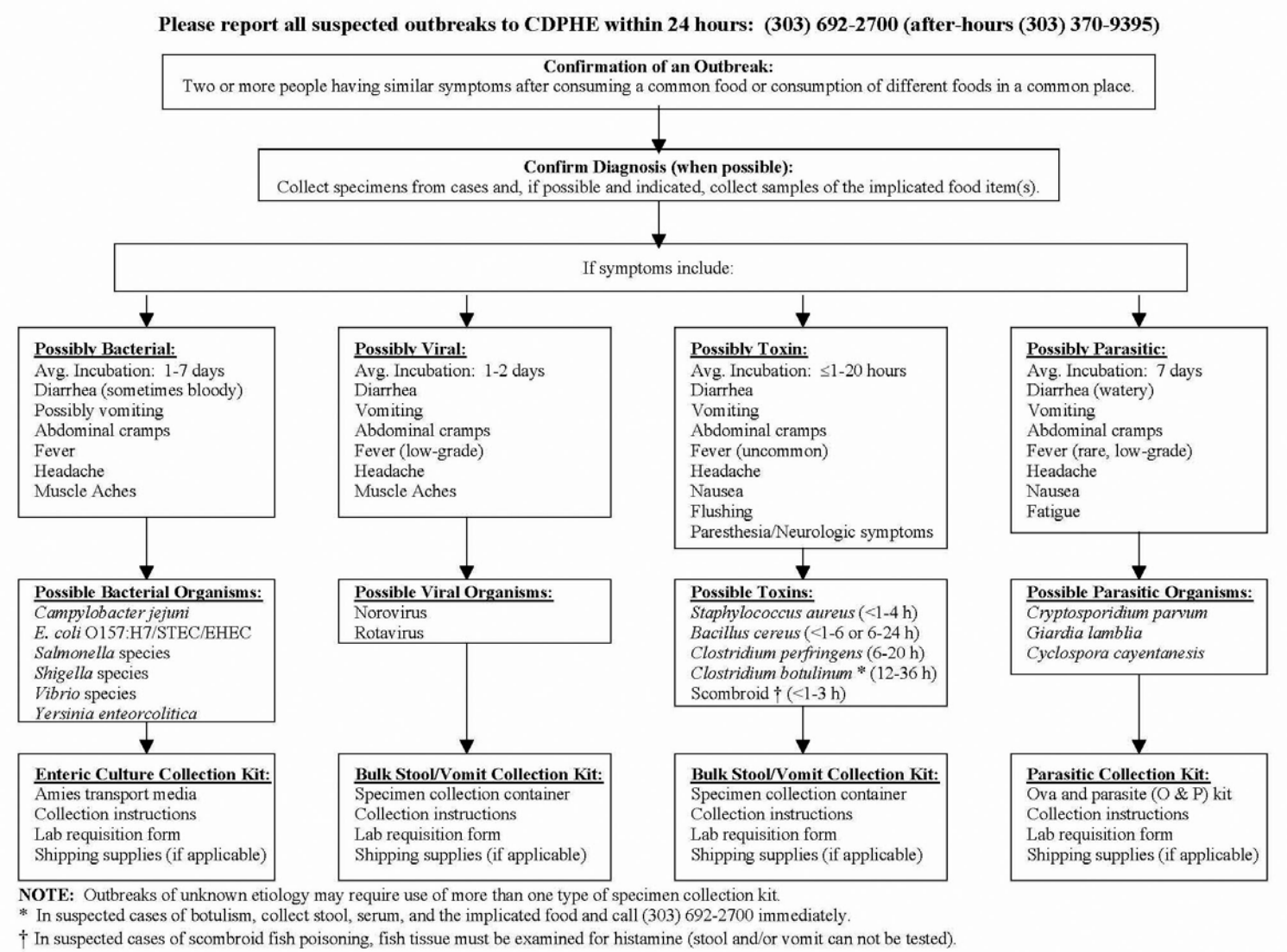
* Direct fluorescent antibody stain ** Polymerase Chain reaction *** Bioassay **** Time-resolved fluorescence (TRF) immunoassay

Time Period for Test Results

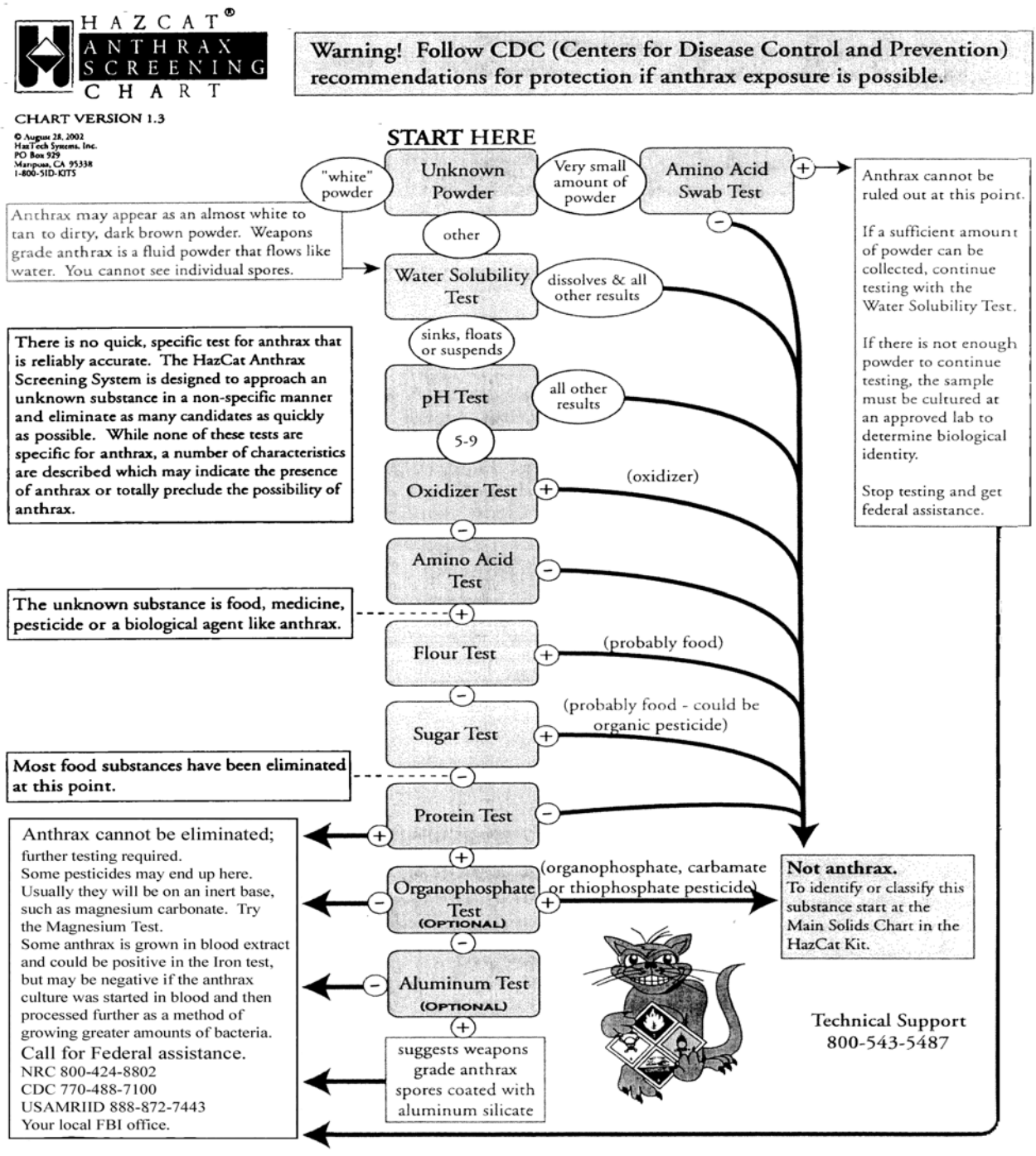
- **Preliminary Testing** (Direct exam, DFA, Antigen, PCR) – **2-4 hours**
- **Confirmatory testing** (culture & identification) – **24-48 hours**

Appendix 5: Outbreak Investigations

Reports of disease outbreaks should be made to CDPHE's Disease Control and Environmental Epidemiology Division. Technical support is available from both the local public/environmental health agency and CDPHE. Samples related to outbreaks should follow an epidemiological investigation must be pre-approved by CDPHE before being brought to the laboratory.



Appendix 6: Anthrax Screening



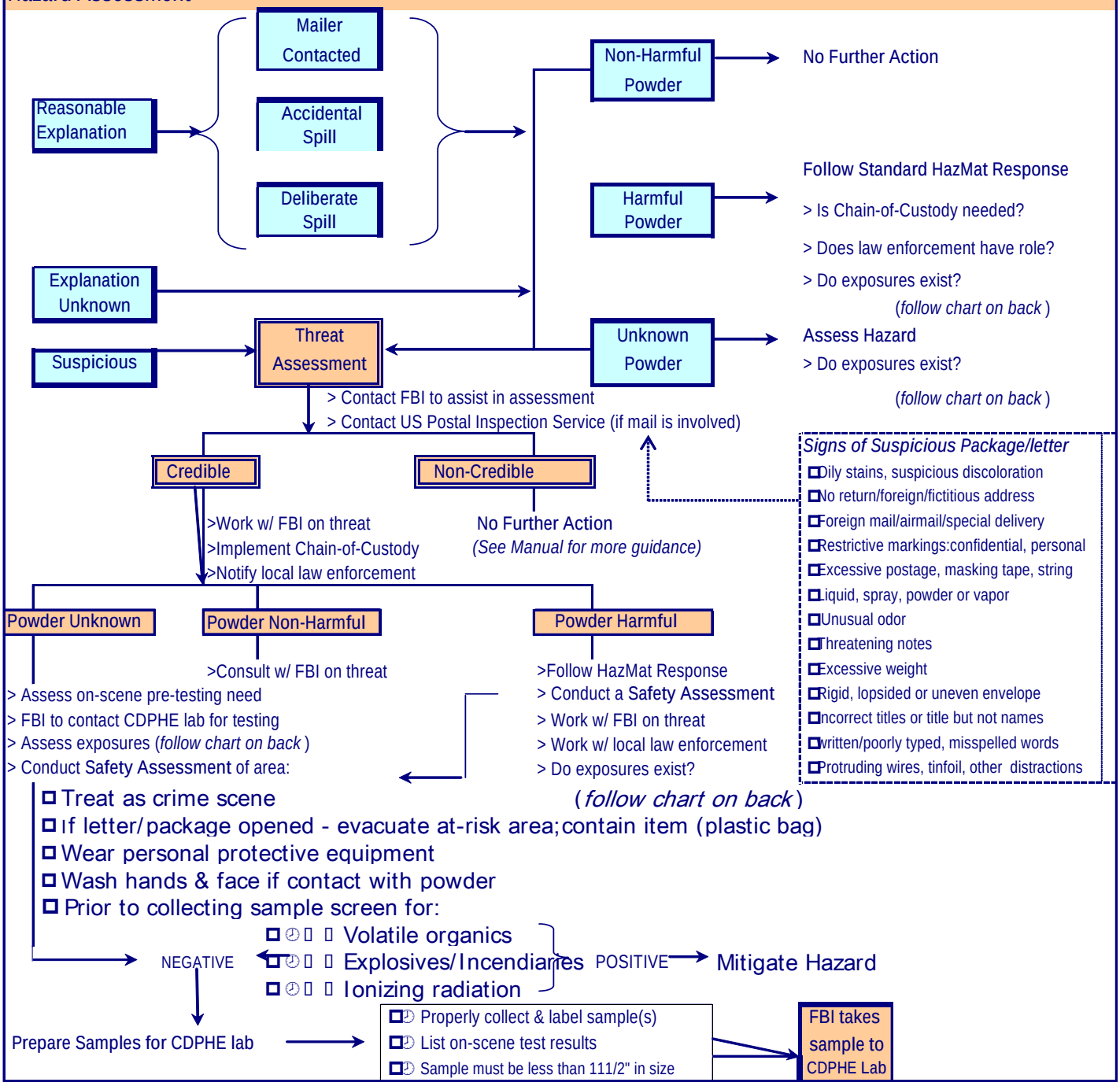
Appendix 7: Suspicious Powder Flowchart

Colorado Department of Public Health and Environment

First Responder Guide: Suspicious Powder Incidents

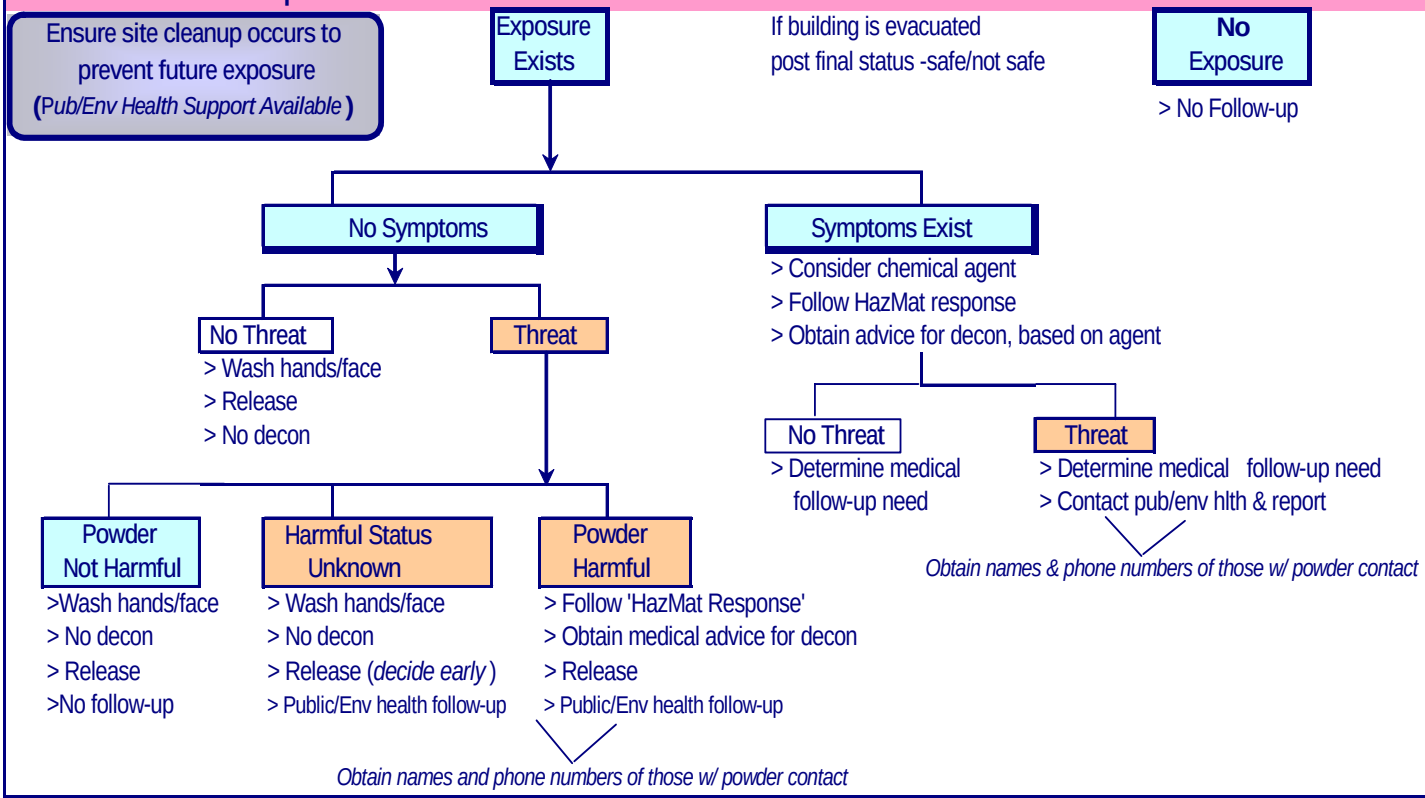
Suspicious powder incidents are a multi-disciplinary response. This document does not replace or supercede any local or agency protocols for primary response or investigations but is intended to compliment the response efforts of those agencies.

Hazard Assessment



The CO Department of Public Health & Environment (CDPHE) created this document in partnership with the CO National Guard Civil Support Team (CST), the U.S. Postal Inspection Service (USPIS) and the Federal Bureau of Investigation (FBI) as a tool to assist responders. 0708

Potential Human Exposures



Important Notification Numbers

Colorado Department Public Health & Environment (CDPHE)	24/7 Emergency Response Line		877-518-5608
	Disease Control Reports	303-692-2700	303-370-9395
Colorado Dept Public Safety	State Patrol Hazmat Response		303-273-1900
	After-Hours		303-239-4501
Colorado National Guard Civil Support Team (CST)	General Assist		720-847-6874
	Request Team		303-279-8855
Federal Bureau Investigation (FBI)			303-629-7171 WMD Coord
U.S. Postal Inspection Service (USPIS)			877-876-2455 opt'n 2
Rocky Mountain Poison and Drug Center	24/7 Denver-Metro Area		303-739-1123
	Outside metro Area		800-222-1222
National Response Center	24/7 Notification		800-424-8802
Regional Public Health Laboratories	Denver County		303-436-7365
	El Paso County		719-578-3121
	Mesa County		970-245-7800
	Pueblo County		719-583-4318
	Weld County		970-304-6415 x2273

Estimated Support Response Time - Credible Threat

Agency	Time to Scene		Time Assessment/Testing	Total time
USPIS	Within 1 hr to scene	(most locations)		
FBI	Within 1 hr to scene		Threat investig'n: 1-3 hrs (w/ local law enf)	
CST	Within 30 min to mobilize	(1-4 hrs to scene)	Preliminary tests: 1-2 hrs (CDPHE to confirm)	
CDPHE Lab	24/7 for sample approval		Bio & Chem tests: 3hrs (after sample arrive)	
				6 hours *(per incident)*

This Guide is taken from the *"First Responder Manual On All-Hazard Environmental Incidents Technical Support & Sampling"* published by the Colorado Department Public Health and Environment, Laboratory Services Divisions and Emergency Preparedness & Response Division (cdphe2005/Rev2008)