CHAPTER 8

Collection, Storage and Transport of Samples for Investigations

A Public Health Inspector's duties, in connection with food hygiene and safety of drinking water, often involve obtaining and transporting of samples for chemical/ microbiological analysis. Adherence to the correct method of obtaining samples, storage, labeling, and conveyance to the laboratory, are all very essential as legal proceedings could commonly ensue. Therefore it is very important to pay attention to all details of the correct procedures that have to be followed.

8.1 Samples for microbiological investigations

- **Water Samples**

Samples of water should be collected in sterile bottles which could be obtained from the Food and Water Microbiology Laboratory, and should be taken care to prevent contamination when collecting the sample. For sampling, chlorinated water bottles containing Sodium thiosulphate should be used, which can also be obtained from the same laboratory.

Collection of a water sample from a tap

1. Wipe the outlet of the tap with a clean cloth to remove any dirt.
2. Wash your hands with soap and water.
3. Open the tap and allow the water to run for 30 seconds.
4. Flame the mouth of the tap for 30 seconds. If the tap is plastic clean the mouth with a cotton wool swab soaked in surgical spirit or 70 % alcohol, and leave for 2 minutes.
5. Open the tap and allow water to run out for 30 seconds.
6. Remove the craft paper covering the lid and open the bottle.
7. Fill 3/4 of the bottle with running water while holding the lid downwards.
8. Replace the lid and the craft paper.

Collection of a water sample from a well

To collect the water sample from the well, use a clear, colourless glass bottle (preferably wide-mouthed) of approximately 500 ml capacity. It should have tight-fitting glass lid or a screw-cap type metal lid. After tying one end of a twine thread (long enough to lower the bottle into the well) around the neck of the bottle, boil the bottle along with the thread, for a period of 10-15 minutes. Now lower the sterile bottle into the well, taking care to avoid contamination the bottle, and collect a sample of water from the centre of water surface, about 1 foot below the surface. About ¾ of the bottle should be filled with water. After withdrawing the bottle with water, close the bottle and wrap around the lid and mouth, with a craft paper (refer

the circular on collection of water samples for analysis, issued by the Ministry of Healthcare & Nutrition, in 2009).

Collection of a water sample from a stream

1. Walk towards centre of the stream as far as possible.
2. Collect the sample, holding the bottle with its mouth directed towards the current of water, about one foot below the surface of water.

After collecting the sample from the tap, well or stream, the label duly filled should be pasted on the bottle. A properly filled request form should accompany the bottle when sending it to the analytical laboratory.

Transport

Transport the sample to the laboratory in a cold box with ice packs. The sample could be transported without ice, if it is possible to transport it within 2 hours of collection. The sample should be sent to the laboratory within 24 hours of collection and must be kept in the refrigerator until dispatch.

- **Food Samples**

General guidelines on collection of food samples

1. Sampling should be carried out by an experienced, trained person. Care should be taken to prevent contamination during collection.
2. Whenever possible samples should be sent to the laboratory in the original, unopened containers or packages.
3. If the products are in bulk, or in large containers, representative samples should be collected using sterile sampling apparatus, into sterile containers. The containers should be wide mouthed, leak-proof and dry.
4. Collect at least 100g of the food item for each sample unit.
5. No preservatives should be added to the samples.
6. Label the sample and send to the analytical laboratory, with a request form duly filled.

Storage and Transport and of food samples

Adequate precautions should be taken to prevent any change occurring in the original microbial flora of the food.

1. Samples should be transported to the laboratory as quickly as possible.

2. Original storage conditions should be maintained during storage and transport.
   - Dehydrated foods, dry foods and canned foods, that are not perishable should be stored and transported without refrigeration. These should be stored in a clean, cool, dust-free place.
   - Refrigerated foods, perishable foods and samples drawn from original packaging and transferred to new containers, should be stored and transported in cold boxes with ice packs. Refrigerated foods should not be frozen.
   - Frozen foods should reach the laboratory in the frozen state. Collect frozen foods in pre-chilled containers (containers kept in the freezer long enough to chill them thoroughly).

Please contact Food and Water Microbiology Laboratory before sending samples. These samples should preferably reach the laboratory during the first 3 days of the week, unless urgent.

- **Clinical Samples**

General guidelines on collection and transport of samples for microbiological investigations;

1. Use sterile containers obtained from the laboratory. These should be leak-proof containers, preferably screw-capped.
2. Avoid contamination when collecting the samples.
3. Samples should be collected before antibiotics treatment is started.
4. A label, containing following information should be pasted on the container.
    - Name of the patient
    - Reference number
    - Investigations required
    - Date (of collection)
5. Samples for Microbiological examination should be sent to the laboratory as soon as possible.
6. A request form, duly filled with following information should accompany the sample;
    - Name, reference number, age, sex, hospital and ward, or address (if not hospitalized)
    - Date and time of collection
    - Investigations requested
    - Clinical history/ reason for requesting the investigations
    - Treatment given, if any
    - Signature, name, designation, contact number (tel /fax) and address of the person sending the sample

    Specimens should be packed well to prevent leakage and breakage during transport.
7. Make communication with the laboratory (before and after sending sample),

    - To seek advise on collection and transport of samples
    - To inform about sample/s to be dispatched to the laboratory
    - To inquire about the report
    - To seek assistance in interpretation of the results of investigations

Stool samples from patients with suspected gastroenteritis/ food poisoning

- A specimen of stools is always better than a rectal swab. A rectal swab should be sent only when it is not possible to obtain a sample of stools.

- A fresh specimen should be sent, if requesting for amoebae - ova and cysts (AOC).

1. Instruct the patient to pass stools into a clean, dry, disinfected bedpan or on to a clean paper.

2. Transfer a portion of stools, preferably from an area containing blood, pus or mucus, into a clean, dry, wide-mouthed and leak-proof container.

3. Transport the sample to the laboratory as soon as possible. Should there be a delay, send the sample in a transport medium such as Carey Blair medium, VR medium. Alkaline Peptone Water (APW) could be used if cholera is suspected.

Samples sent for the investigation of food poisoning

- Samples of remnants of implicated foods and stools and vomitus of patients could be sent in sterile and wide mouthed containers.

- The request form accompanying samples should contain relevant data such as symptoms, time interval between consumption of implicated food and onset of symptoms.

Stool samples from a patient having Acute Flaccid Paralysis (AFP)

It is mandatory that all patients with acute flaccid paralysis must be reported to the epidemiologist, and subjected to virological examination to exclude polio.

- Two stool samples each should be collected from all patients with AFP, within 14 days of onset of paralysis. As the virus concentration decreases with time all attempts must be made to collect the samples very early in the infection.

- As the viral shedding is intermittent, the two stool samples should be taken 24-48 hours apart.

- The quantity of a sample should be 8-10 g each (size of two adult thumb nails or two tamarind seeds).

- Stools should be collected in a clean, preferably sterile, screw capped, leak-proof bottle or in a container provided by the Epidemiology Unit, Colombo.

- Labeled samples should be sent to the laboratory with a request form containing date of onset of paralysis, date of collection of stools, date of dispatch of stools and date of the last polio vaccination dose taken by the patient.

- Stool samples have to be carefully sealed in containers and stored immediately in a refrigerator or packed in a cold box with ice packs, pending transport. Undue delays or prolonged exposure to heat may destroy the virus. Sample should reach the laboratory within 72 hours of collection.

- The samples should be hand delivered to the MRI in a cold box packed with ice. Samples are received at the MRI throughout the day and on all days including holidays. The samples should be handed over to the MLT at the polio laboratory or to the Relief MLT (RMLT) on duty, if it is outside duty hours.

Sputum for diagnosis of Tuberculosis

- When suspecting pulmonary tuberculosis, three sputum samples should be sent for the detection of Acid Fast Bacilli (AFB) and culture.

- Sputum samples should be collected in the open air, away from other people. If a room is used it should be a separate and well ventilated room.

- Sputum samples should be collected into clean, wide mouthed, leak-proof, unbreakable containers with tight-fitting lids, preferably screw capped. The container should be easily disposable by burning, after the necessary smears are made and the culture media are plated.

Procedure of collecting sputum into the container

- Explain to the patient, the reason for sputum examination
- Demonstrate how to open and close the container
- Give the labeled container to the patient and explain how to collect the sample

The sample should be an expectorated sputum and not saliva. Advise the patient to deeply inhale 2-3 times and cough out sputum deeply from the chest, into the container held close to the mouth. 3-5 ml of good quality (thick) sputum should be collected without contaminating the outside of the container.

- Close the container tightly
- If there is a delay in transporting, the samples should be kept at 4^0 C, to retard the multiplication rate of commensals.

Sputum samples are best collected in early morning as soon as the patient wakes up, if the sample is collected at home. Advise the patient to rinse his/her mouth, before collecting the sample.

Head of a suspected rabid dog

The head separated at the neck should be put into a container with a tightly fitting lid and then this container is put in a bigger container with ice. Specimen is to be labeled and hand delivered to the MRI. Animal heads are received at the MRI throughout the day, on all days including holidays.

Blood films

For the detection of Malaria Parasites
A thick and a thin smear should be prepared on the same slide.

The pulp of the finger should be pricked with a sterile lancet or needle after cleaning the skin with 70% alcohol. The first drop of blood is wiped away with cotton wool, and the bleeding finger is touched with one end of the surface of the slide and three drops of blood deposited. Another drop is taken to the same surface of the slide inner to the area where the three drops of blood are. Then with the clean unbroken edge of another slide, the single drop of blood is spread uniformly into a thin smear. The three drops of blood taken first are then smeared to cover an area of the size of a 25 cts coin.

For the detection of Filaria Parasites
Only a thick blood film prepared as above is needed.

Dispatch of blood films
Blood films should be air dried. Each film should be wrapped separately with tissue paper or separated from each other by keeping match sticks or ekels in between so that they will not stick to each other.

8.2 Samples for Chemical Analysis

Food sampling
Sampling is an important aspect when sending them for laboratory analysis.

Why food samples are analyzed?
To check

- The purity of food
- the product quality
- contamination
- Adulteration
- Composition

Why food samples are collected?

- Consumer complaints
- Asses the violation of existing regulations
- Examine the products in the market
- Preventive action

Who are the authorized officers to collect food samples?

- Medical Officers of Health
- Food and drugs Inspectors
- Public Health Inspectors
- Veterinary surgeons in relation to examination and seizure of meat

What are the types of samples?

- Formal
- Informal

What does formal and informal sampling means?

- Informal samples are taken for surveillance and monitoring purposes and to provide advice to food manufacturers
- Formal samples are taken for the purpose of legal evidence as laid down in the Food act No. 26 at1980 or is sent to an approved laboratory

Where the samples are collecting from?

- Processing line
- Transport
- Storage warehouse
- Retail shelf

What are the forms of food samples?

- Solids e.g. Rice
- Liquids e.g. Coconut oil
- Semi solids e.g. ghee
- Solids in liquid e.g. canned fish

How do we collect a sample for analysis?
Laboratory sample should be a representative sample of the whole food sample.
The following steps should be taken when collecting a proper sample;

(i) *Selective sampling* – If there are visible contaminants or adulterants, collect the sample directly from such containers.

(ii) *Random sampling* – If there are no visible contaminants and adulterants random sampling is possible.

During random sampling, the number of containers available for sampling at the ware house, or at bulk storage and the type of food item must be considered. The sampling should be according to the SLS standards (SLS 428: 1977 - Random Sampling Methods)

E.g- Iodated salt – The number of containers to be selected from each lot shall be in accordance with the following table.

Scale of sampling

Lot size	Number of containers to be selected
Up to 150	4
151 to 280	7
281 to 500	10
501 and above	16

Containers shall be selected at random, by making use of the 'Table of random numbers' as given in **SLS 428; 1977,** to ensure proper selection.

Points to be considered when submitting a food sample for analysis

1. Cereals, Pulses, whole spices and all powdered samples should be wrapped in polythene or polypropylene (do not use paper as wrapping).

2. Enclose the sealed sample in a suitable cover and seal latter also properly. Label the sample clearly including the following information:-

    - Name of the PHI / F & D I
    - Area
    - Sample number
    - Date and time of sampling (time is important for microbiological analysis)
    - Common name (generic name) of the food item and type of the sample (formal/ informal)
    - Local authority

A letter requesting analysis should be submitted with each sample, containing the following information;

- Common name (generic name) of the food item
- Type of sample (formal/ Informal)
- Sample number
- Date and time of sampling
- Name of the PHI / F & DI
- PHI area

- Local authority area
- Address of PHI / F & DI
- Any other complaints like whether the stock seal was there or not, consumer complaints
- Special tests to be carried out

3. Submit the sample as soon as possible after the collection.

4. Liquid samples should be sent in glass or plastic bottles. Never use polythene bags.

5. Do not use glass containers when sending samples by post.

6. Frozen foods (ice cream, ice lollies, yoghurt etc) should be sent immediately in frozen state (0° C). This ensures that frozen samples are received at the laboratory in their original state. Vaccine carriers or ice bags can be useful.

7. Bread samples must be sent immediately after collection as the moisture analysis should be completed within 24 hours.

8. When sending preservatives (e.gs. Cordials, Jams) for analysis, the original seals on the bottle should not be disturbed.

9. Submit a reasonable quantity of the food sample, sufficient for duplicate analysis to be carried out.

10. Memorandum published in the Govt. gazette of 07.06.85 shall be submitted with the samples.

Specimen form of a label to be attached to the sample

Food Sampling under the Food Act No. 26 of 1980

Pradesheeya Sabha, Municipal/ Urban council…. ..……

Chemical analysis/ Microbiological examination

1. Type of sample: Formal/Informal
2. Name of the sample:
3. Reference number of the Inspector:
4. Date and Time of sampling:
5. Name and address of the vendor: (optional)
6. Whether preservatives are added? : yes/no
7. Name of the authorized officer:
8. Area of the authorized officer:
9. Any special reasons for sampling :

Signature of the authorized officer

Signature of the vendor

Specimen Form of a Requisition to be sent to the approved analyst / additional approved analyst, with the food sample

To the Approved analyst /Additional approved analyst
From the PHI/ F& DI of …… of ……Pradesheeya Sabha / Urban council / Municipal Council

The following sample is submitted herewith for chemical analysis/ microbiological examination, under the Food Act No. 26 of 1980.

Please send the analyst Certificate / Report to the following address.

(State your address) ----------

i. Type of sample – Formal/ Informal
ii. Name of the sample:
iii. PHI reference number:
iv. Date and Time of sampling:
v. Name and address of the vendor: (Optional)
vi. Whether any preservatives for insect contamination (formaldehyde) are added? : yes/ no if yes; quantity added:
vii. Please indicate any special tests to be carried out (infestation/moulds/extraneous matter/colouring matter/artificial sweeteners/suitability for consumption etc.)
viii. Lot size of the food article seized under the Food Act No. 26 of 1980 (actual quantity should be declared):
ix. Specimen seal of the actual seal on the sample:

Vendor's specimen seal

authorized officer's specimen seal

Vendor's signature/thumb impression
……

……..
Name and signature of the authorized Officer

Sample quantities required for chemical analysis of food

	Food Item	Minimum quantity required
1.	Baking powder	50g or the original pack
2.	Bee's honey and treacle	50ml
3.	Bread	1 loaf
4.	Butter, cheese, margarine and ghee	100g
5.	Cereals and pulses	250g
6.	Chocolate	50g (container sealed)
7.	Fish, Dry Fish	250g
8.	Foods made out of flours and cereals (papadam, biscuits, etc.,)	100g
9.	Fruit Juices / Soft Drinks	200ml (for preservative Testing)
10.	Ice – cream	150ml or 2 cups

11. Ice Lollies / Ice packet	200m
12kij. Iodized Salt / Salt	100g (original pack required)
13. Jam/Jelly	100g (original pack required)
14. Milk	200ml
15. Milk Powder	250/400g (original pack required)
16. Milk Toffees	200g
17. Oils	100ml
18.Spices (whole / powdered)	100g
19. Sweets	100g
20. Tea, Coffee, and Cocoa	100g
21. Vinegar	375ml
22. Water	2.5 l
23. Yoghurt	150ml or 2 cups

Please Note

a. Sending a sufficient quantity of food material will help to carry out duplicate analysis and confirmatory tests.
b. Cereals, pulses, whole condiments and all powders should be wrapped in polythene or polypropylene(do not use papers).
c. Label the sample clearly, with the name of the PHI, area, Local Authority address etc.
d. Laboratory sample should be a representative sample of the whole food sample.
e. Liquid sample should be sent in glass or plastic bottles.
f. Frozen Foods (Ice cream, Ice Lollies, Yoghurt etc.) should be sent immediately after collection, under frozen condition (below zero degrees centigrade),so that frozen samples are received at the laboratory, in the original form.
g. If preservatives are to be tested, send samples intact in their original container.