Chapter 7: Sampling

Going out and collecting a sample without first doing some planning may result in a meaningless, contaminated, or unrepresentative sample. No matter how precise lab techniques are, if the sample being testing has not been obtained, stored, or preserved in the proper manner, the results may be invalid and time and material used to test the sample will have been wasted. The greatest lab technician in the world will not obtain meaningful results from a poorly-collected sample. If the results are to be meaningful—that is, if the data is needed to indicate the conditions present when the sample was taken, then some fore-thought to sample collection is necessary. There are many questions to consider before actually collecting a sample. The answers to these questions will help put together a sampling plan which is easier and more effective.

Prepare a Sampling Plan

A sampling plan should address the following questions.

1. Why is the sample being collected?
2. What tests need to be run on the sample?
3. Where is the sample going to be collected from?
4. How is the sample going to be collected?
5. When does the sample need to be collected and analyzed?

Why is the sample being collected?
Wastewater samples are collected for a variety of reasons.

1. Federal and State Regulations
2. Process Control
3. Plant expansion
4. Surcharges

In 1972, Congress passed PL 92500 which became known as the "Clean Water Act". This act provided money for the construction of wastewater treatment plants, operator training, and research. The money had strings attached which required municipalities to monitor the quality of their effluent. As a result a permit system was established by EPA which required municipalities to self-monitoring the quality of their effluent. The NPDES (National Pollutant Discharge Elimination System) system was established and municipalities were required to submit DMRs (Discharge Monitoring Reports). Laboratory tests such as BOD, TSS, pH, and Fecal Coliforms were common to all DMRs.

WWTP operators/managers also run laboratory tests to monitor treatment plant operation. These tests are called process control tests and indicate how well a particular process is working. Lab personnel and/or operators will take samples and run tests to make operational decisions. For example, in a typical wastewater treatment plant there are several places to sample. As wastewater flows through the treatment plant (and through the collection system) its characteristics change. By taking samples at the different treatment basins, information can be gathered to help decide how the plant is operating and whether operational changes need to be made to improve treatment. If sampling and testing procedures are correct, the lab technician can use the test results combined with plant observations and knowledge of the plant operational procedures to formulate a fairly accurate picture or pattern of what is happening in the plant. Operational questions such as:

1. Is the dissolved oxygen level in the aeration basins adequate?
2. How often and how much sludge do you want to waste?
3. Is the chlorine dosage sufficient for disinfection without having too high a residual?
4. Are solids settling well in the primary clarifier?
5. Is the aeration basin removing the BOD effectively?
6. Is there a oil and grease problem in the collection system?

can be answered through laboratory analysis. Timely information gathered from test results will help answer these and other questions.

Laboratory data can also be collected for planning purposes. Are you sampling specifically to justify a new budget or a new procedure or a plant expansion? Collecting laboratory data over a period of time can provide plant managers and operator sufficient evidence to justify building additional treatment units, replacing old units, etc.

Municipalities are often only concerned with the wastewater that arrives at the headworks to the WWTP. Eventually it becomes obvious that the waste contributors must be identified. Larger municipalities establish IPP (Industrial Pre-treatment departments) with full time staff responsible for collecting samples from industry. Industrial samples can play a major role in WWTP operation. Meat packers, food processors, beverage producers, semiconductor manufacturers, etc can all discharge high volumes of high strength, high volume, or toxic materials. Industries are often charged extra when they discharge abnormal amounts. This extra charge is called a surcharge. For example, a coca cola bottling plant may discharge a high sugar waste (BOD of 10000 mg/L). This high level of waste will require additional treatment capacity at the WWTP. If a normal consumer discharges a BOD of 100 mg/L, the industry will be billed more than a normal consumer. Laboratory data will justify the surcharge.

What specific tests need to be run on the sample?

Knowing what is going to be sampled will help determine the locations and the number of sampling sites necessary to obtain all the information you want.

A single collected sample can be used for several different tests. Knowing the reason for sampling helps determine what types of tests need to be run and how much sample to collect. Being familiar with the various testing procedures allows the technician to be able to estimate the volume of sample that needs to be collected. Knowing what tests needs to be run also helps budget technician time. Time management is a skill that all lab technicians will have to develop. Lab technicians will often be running several tests at the same time. In order to do this proficiently, the lab technician must have a work plan, taking into account holding times, preservation, drying times, etc. The lab technician may also have to prepare chemicals or reagents ahead of time. Other chemicals may need to be prepared the day of the test because they are unstable and quickly deteriorate in quality.

Knowing the specific tests required also requires the lab technician to understand the wastewater characteristics. Special sampling methods or preservation techniques may be needed. If you are sampling for dissolved oxygen at various depths in a basin, reservoir or lake, there are certain sampling procedures required so that your oxygen levels will be accurate. If the sample contains chlorine, and bacteriological tests need to be performed, a dechlorinating agent will need to be added to remove the excess chlorine so that further disinfection does not occur after the sample has been collected. If samples are very cloudy or turbid, you may want to use a different procedure for some tests to minimize the effects of the turbidity. Refrigerating samples (1 - 6°C) will lessen the chance that bacteria will either die-off or multiply, thus ensuring more accurate results from a bacteriological test such as fecal coliform.

Where are the samples going to be collected?

If samples are collected from the collection system, which manhole will be used? Does the technician have the necessary collection equipment? Is there a traffic concern? Is there a toxic environment concern? Will additional manpower be needed? Will the sample be well mixed at the sampling point?

If the sample is collected within the treatment plant, the technician should consider the purpose of the treatment unit. Where should TSS samples be collected? Where should BOD samples be collected? Do you collect the sample from the supernatant? Do you collect the sample from a well mixed area? Where is the best place to collect a sample for microscopic examination of the aeration basin?

How is the sample going to be collected? Are you going to obtain a grab sample or a composite sample?

Grab Sample

A grab sample consists of a single container or bucketful of wastewater that is "grabbed" at one specific time. Grab samples indicate the condition of the wastewater at that specific time and may or may not represent the normal conditions. Grab samples are required when the contaminant concentrations change rapidly. For instance, grab samples are required for certain tests, such a pH, D.O. (dissolved oxygen), and bacteriological analyses.

Composite Sample

A composite sample consists of several grab samples collected from the same spot over a specific period of time and merged into a single sample. A composite sample is more arduous, complicated and usually inconvenient than a simple grab sample. Collecting a sample every few minutes and adding it to a single bottle is tedious, boring, and costly. To help solve this problem, a 24-hour automatic sampler is often used. The automatic sampler consists of a battery pack, a programmable timer, a pump, and as many as 24 bottles.

The automatic sampler can be programmed to draw a certain volume of sample every few minutes and deposit each sample into one of the iced (preserved) bottles.

At the end of the sampling period, the lab technician can retrieve the bottles, bring them back to the lab and create a single composite sample. Analysis can now be performed on a single composite sample that is more representative of the wastewater quality than a grab sample. ***SAFETY: Be sure to handle the automatic sampler safely. A full sampler can weigh as much as 80 pounds.***

Unweighted composite

An unweighted composite consists of collecting the same sample volume at a constant time interval. For instance, the technician collects 400 ml every 30 minutes for 6 hours. At the end of the time period, there will be 12 individual bottles representing the wastewater quality over the 6 hour time period. The technician now composites the samples by pouring say 200 ml from each bottle into a large 1 gallon plastic bottle and mixes. The technician now has 2400 ml of a 6 hour composite sample that can be used for analysis. Unfortunately, the sample at 7 am was collected when the water was pretty clear and hardly flowing while the sample at 10:30 was collected when the wastewater was very cloudy and flowing very rapidly. Both these sample are given the same proportion in the composite yet the 10:30 sample represented a higher percentage of the actual wastewater flow. To reduce this unweighted effect, a flow proportional or flow weighted composite is collected.

Flow weighted composite

A flow proportional composite is collected using a flow meter connected to the automatic sampler. The flow meter will measure the flow in the wastewater stream. As the flow increases during the day, more samples are collected, thereby collecting a greater volume at the time

when the flow is highest. Using the same example as before, the technician sets the automatic sampler to collect 20 ml every time the wastewater total flow increases by 10,000 gallons. At 7:52, the flow totalizer goes over the 10,000 gallon mark, the sampler is triggered to collect 20 ml and places it in the 1 gallon container in the center of the automatic sampler. At 8:42, the totalizer now records the second 10,000 gallons for the day and again collects 20 ml. As the daily flow increases, the number of samples collected and composited increases. Eventually, at 10:30, the sampler may be collecting samples every 10 minutes. At the end of the 6 hour composite period, the samples in the 1 gallon container represent the composite sample and the technician can begin the analysis immediately.

For labs that do not have sampling flow meters, the waste treatment plant flow totalizer log can be used. In this instance, the automatic sampler will be triggered by time rather than flow. For instance, the sampler was started at 7 am and set to collect 80 ml every ten minutes. At 7:10, 80 ml will be delivered into bottle #1, at 7:20, a second 80 ml will be delivered to bottle #1, again at 7:30, 7:40, 7:50 and finally at 8:00. Bottle #1 will now contain a total of 480 ml. This process will repeat again but the sample will now be added to bottle #2. At the end of the 6 hour composite period, the automatic sampler will contain 6 bottles each containing 480 ml. The technician must now composite the sample by using the WWTP flow meter to determine how much to removed from each bottle.

The formula is:

$$\frac{\text{Flow at time} \times \text{volume of sample needed}}{\text{Total flow during composite period}} =$$

General composite procedure:
1. Estimate the total amount of composited sample that will be needed for analysis. Be sure to collect extra in case of spillage. Doubling the amount needed is a nice safety factor. For instance, the lab needs about 2 liters to run BOD, TSS, and TKN. The technician decides to collect about 4 liters.
2. Collect the samples.
3. Record the flow readings for each time a sample is taken.
4. Add up the flows (it really does not matter what the units of flow are; what is significant is how the flows are proportional to each other)

For example, determine how much sample should be composited from each bottle. Assume 2000 ml should be collected for the analysis

Time	Flow, gpd	Volume in bottle	Volume to composite
8:00 am	40,000	880 ml	
9:00 am	80,000	880 ml	
10:00 am	100,000	880 ml	
11:00 am	200,000	880 ml	
12:00 pm	120,000	880 ml	
1:00 pm	60,000	880 ml	

Answer: The total flow is 600,000 gal. Using the formula, the amount of sample that should be removed from bottle #1 (8:00am) is:

$$\frac{40{,}000 \text{ gpd} \times 2000}{600{,}000 \text{ gpd}} \text{ ml} = 133 \text{ ml}$$

Calculate the amount that should be composited from the remaining samples. Notice that more sample is composited during the higher flows.

Time	Flow, gpd	Volume in bottle	Volume to composite
8:00 am	40,000	880 ml	133 ml
9:00 am	80,000	880 ml	267 ml
10:00 am	100,000	880 ml	333 ml
11:00 am	200,000	880 ml	667 ml
12:00 pm	120,000	880 ml	400 ml
1:00 pm	60,000	880 ml	200 ml

When these volumes are added to the sample container, the final volume will be 2000 ml.

Mix the sample well before analysis. BOD and TSS are often performed on composited samples.

There is another type of composite sample, which involves sampling at different locations such as a compost sludge pile, rather than sampling over a period of time. For example: sludge regulations require that pathogens be destroyed prior to disposal. A composite sample of the sludge is collected by grabbing samples from various areas of the sludge pile then composite them.

When does the sample need to be collected and analyzed?

Finally, the lab technician needs to know when the sample should be collected and how long the sample can be held before analysis must begin.

For instance, the discharge permit may require the sample be collected over the peak flow period. If the sample is to be a 6 hour composite, the lab technician may need to have the sampler prepared by 8:00 am to be able to collect the composite over the 6 hour peak flow period. Will there be enough time to composite and analyze the sample? In this example, the composite will finish at 14:00. How long will it take to prepare the composite? How long does the analysis take? Can the sample be held until the next morning? The lab technician needs to plan? If the sample cannot be preserved overnight, has the lab technician done preliminary preparation to expedite the analysis?

Sample Collection

Safety First!!! Laboratory safety was discussed earlier, but many of the same precautions are required when collecting wastewater samples. Gloves, goggles, and lab coats are the minimum requirements for a safe sample collection plan. Avoid selecting sampling sites that present a hazard such as leaning over railings, slippery decks, etc.

Prepare the sample bottles

Choose sample bottles that will not affect or react with the sample. For most normal wastewater samples, either glass or plastic is acceptable. The most common glass container is composed of borosilicate glass (hard glass). Borosilicate glass is marketed as Pyrex or Kimax. Plastic bottles are generally composed of LPE (Linear Polyethylene) or PP (polypropylene). There are many other types of sampling containers and the sampler should be aware of the composition when analyzing for trace contaminants. Containers may decompose and release contaminants or sorb materials onto the walls of the bottle. The sampler should also be aware of the type of bottle cap used. For instance, caps with metal foil liners should not be used when collecting metal samples. For trace analysis, the type of sample container should be documented.

For microbiological analysis such as total or fecal coliform, the sample bottle must be sterile and contain a dechlorinating agent such as sodium thiosulfate. The minimum bottle size is 120 ml and at least 1" of air space must be present to allow for adequate mixing.

Use amber colored bottles for collecting light sensitive materials, usually organics.

Discard cracked, chipped and etched glass bottles. Discard crazed plastic bottles. Discard bottles which are leaking.

Just like lab glassware, sample bottles should be clean and dry. Clean by using (alconox) detergent and hot water; rinse with tap water 10x to remove all trace of detergent; then rinse five times with small amounts of DI water. In most cases, the sample container should never be rinsed with the sample prior to collection. Pre-rinsing may allow some contaminants to stick to the sides of the container or settle to the bottom prior to the final sample collection. For instance, pre-rinsing with an influent sample

may cause "extra" oil and grease to stick to the sides or "extra" settleable solids to remain in the bottom of the bottle. This would cause a high bias to the sample answer. Pre-rinsing may also rinse out pre-loaded preservatives such as acid. Samples collected for total or fecal coliforms should never be rinsed. Rinsing will remove the dechlorinating agent.

Site Selection

Remember, the test result is only as good as the sample collected. Select a sample site that is

1. safe
2. representative
3. uncontaminated.

Sample Identification

Once the sample is collected it must be labeled. Label each sample container with an adhesive tag or label. Know in advance what information you will put on the container. An information form preprinted on the tags or labels provides uniformity of sample records, assists the sampler, and helps ensure that vital information will not be omitted. Useful information to put on the sample container includes:

1. Sample identity code.
2. Signature of sampler.
3. Signature of witness.
4. Description of sampling location detailed enough to accommodate reproducible sampling. (It may be more convenient to record the details in the field record book). This may include depth when sampling ponds or clarifyers.
5. Sampling equipment used.
6. Date of collection.
7. Time of collection.
8. Type of sample (grab or composite).
9. Water Temperature.
10. Sampling conditions such as weather, water level, flow rate of source, etc.
11. Any preservative additions or techniques.
12. Record of any test results done in the field.
13. Type of analyses to be done in laboratory.

It's guaranteed, that lab technicians who do not label sample bottles will eventually get the bottles mixed up either in route to the lab or in the lab.

Preservation, Storage, and Holding Times

Deliver the sample to the laboratory as soon as possible after collection. Holding time is considered the time interval between collection and analysis. Samples begin to change as soon as they are removed from their environment. The shorter the holding time the more reliable the analytical results will be. If the sample cannot be delivered to the laboratory within 2 hours, it should be preserved. Preservation only helps retard chemical and biological changes that begin immediately after sample collection. The sampler should plan for transportation to the laboratory. If the sample must be shipped, commercial transportation such as FEDEX ® and bus pickup and delivery schedules must be considered.

Some samples cannot be preserved because the sample conditions change quickly. These samples must be analyzed either in situ or on site immediately after collection. Such analyses would be pH, DO, chlorine residual, and temperature.

There are no absolute methods of preservation but the methods used are generally intended to retard biological action, retard chemical changes and reduce volatility of organic molecules. Ice samples that need to be transported to the laboratory or that will be composited. If using an automatic sampler, the bottles in the sampler should be packed in ice during the compositing period. Do not freeze samples or use dry ice. Once the samples reach the laboratory, refrigeration is used to retard biological activity and reduce volatilization of organics until the time of analysis. Be sure to document time of arrival and refrigeration temperature.

Chemical contaminants such as metals are commonly preserved using acid and dropping the pH to below 2. The addition of acid drops the pH and helps retard chemical oxidation and precipitation reactions. Ideally add the acid prior to the sample bottle so that preservation begins immediately. Check the pH and document at the time of collection.
Once the samples have been delivered to the laboratory, it is the responsibility of the lab manager/technician to maintain the preservation and assure timely analysis. Samples that exceed the holding time should be invalidated or identified as such. For composited samples, it is common practice to use the time at the end of the composite collection as the sample collection time.

Chain of Custody

Most samples are simply collected by the wastewater laboratory technician or operator, labeled and returned to the laboratory for analysis. However, when legal litigation is indicated, it is necessary to have an accurate written record to trace possession and handling of samples from collection through reporting. This chain of custody should be able to identify who handled the sample from collection, to transport, to storage, to analysis, to destruction. An example of a chain of custody form is included in Appendix A

A sample is in "custody" if:

1. It is in one's actual physical possession.
2. It is in one's view, after being in one's physical possession.
3. It is in one's physical possession and then locked up so that no one can tamper with it.
4. It is kept in a secured area, restricted to authorized personnel only.

Collection

Field collection records should be completed at the time the sample is collected and should be signed by the collector. Typical field information would be:

a. Date and time
b. Sample location (be detailed)
c. Preservation used
d. Analysis requested
e. Name and signature of collector(s)
f. Pertinent field data (DO, PH, Chlorine residual, temperature)
g. Serial numbers of transport cases, sample log numbers
h. Comments

The sample should have a waterproof label and the information written with an indelible pen. The sample container should be closed, placed in the transport case, then sealed and transported.

Transport

Usually samples that are headed for court are not sent to the in-house lab, which means they may be hand delivered, delivered by mail, or delivered by common carrier. The chain of custody form must accompany the sample. If hand delivering, have the receiving person sign the form. If sending by mail, send as registered mail with return receipt requested. If sending by common carrier, keep the sending receipt.

Laboratory

Laboratory personnel are responsible for the care and custody of the sample once it is received and be able to testify that the sample was secured in the laboratory at all times from the moment it was received until the time it was disposed. The receiving laboratory will usually designate one person (the custodian) to be responsible for the sample custody. The custodian will maintain a lab book which tracks the movement of the sample throughout the laboratory. The log book will indicate who removed the sample from the custody area, when it was removed, when it was returned, when it was destroyed.

Disposal

The laboratory custodian will dispose of the sample when it is certain that the information is no longer required, or the sample has deteriorated. A sample is considered to have deteriorated when the maximum holding time has elapsed.