CHAPTER 2

Tools of the Trade

Analytical chemists use a range of equipment, from simple glassware to complex instruments that measure spectroscopic or electrical properties of analytes. The object to be analyzed might be as large as a vein of ore in a mountainside or as small as a vesicle inside a living cell. This course should expose you to some of the instrumental techniques of modern analytical chemistry. Along the way, you must gain some understanding and proficiency in "wet" laboratory operations with simple glassware. *The most sophisticated instruments are useless if you cannot prepare accurate standards for calibration or accurate, representative samples of an unknown for analysis.* This chapter describes basic laboratory apparatus and manipulations associated with chemical measurements.

2-1 Safety, Waste Disposal, and Green Chemistry

The primary safety rule is to do nothing that you (or your instructor) consider to be dangerous. If you believe that an operation is hazardous, discuss it with your instructor and do not proceed until sensible procedures and precautions are in place. If you still consider an activity to be too dangerous, don't do it.

Before beginning work, you should be familiar with safety precautions appropriate to your laboratory. Wear goggles (Figure 2-1) or safety glasses with side shields at all times in the lab to protect yourself from flying chemicals and glass. Even if you are very careful, one of your neighbors may be more accident prone. Wear a flame-resistant lab coat, long pants, and shoes that cover your feet to protect yourself from spills and flames. Rubber gloves can protect you when pouring concentrated acids, but organic solvents can penetrate rubber. Food and chemicals should not mix: Don't bring food or drink into the lab.

Treat chemical spills on your skin *immediately* by flooding the affected area with water and then seek medical attention. Clean up spills on the bench, floor, or reagent bottles immediately to prevent accidental contact by the next person who comes along.

Solvents and concentrated acids that produce harmful fumes should be handled in a fume hood that sweeps vapors away from you and out through a vent on the

Figure 2-1 Goggles or safety glasses with side shields are required in every laboratory.

Limitations of gloves In 1997, popular Dartmouth College chemistry professor Karen Wetterhahn, age 48, died from a drop of dimethylmercury absorbed through the latex rubber gloves that she was wearing. Many organic compounds readily penetrate rubber. Wetterhahn was an expert in the biochemistry of metals and the first female professor of chemistry at Dartmouth. She had two children and played a major role in bringing more women into science and engineering.

roof. The hood is not meant to transfer toxic vapors from the chemistry building to the cafeteria. Never generate large quantities of toxic fumes in the hood. If you use a toxic gas in a fume hood, bubble excess gas through a chemical trap or burn it in a flame to prevent its escape from the hood.

Label every vessel to show what it contains. Without labels, you *will* forget what is in some containers. Unlabeled waste is extremely expensive to discard, because you must analyze the contents before you can legally dispose of it. Chemically incompatible wastes should never be mixed.

If we want our grandchildren to inherit a habitable planet, we need to minimize waste production and dispose of chemical waste in a responsible manner.[1] When it is economically feasible, recycling of chemicals is preferable to waste disposal. Carcinogenic dichromate ($Cr_2O_7^{2-}$) waste provides an example of an accepted disposal strategy. Cr(VI) from dichromate should be reduced to Cr(III) with sodium hydrogen sulfite ($NaHSO_3$) and precipitated with hydroxide as $Cr(OH)_3$. The solution is then evaporated to dryness and the solid is discarded in an approved landfill that is lined to prevent escape of the chemicals. Wastes such as silver and gold that can be economically recycled should be chemically treated to recover the metal.

Green chemistry is a set of principles intended to change our behavior in a manner that will help sustain the habitability of Earth.[2] Examples of unsustainable behavior are to consume a limited resource and to dispose of waste in a manner that poisons our air, water, or land. Green chemistry seeks to design chemical products and processes to reduce the use of resources and energy and the generation of hazardous waste. It is better to design a process to prevent waste than to dispose of waste. If possible, use resources that are renewable and generate waste that is not hazardous. For analytical chemistry, it is desirable to design analytical procedures to consume minimal quantities of chemicals and solvents and to substitute less toxic substances for more toxic substances. "Microscale" classroom experiments are encouraged to reduce the cost of reagents and the generation of waste.

2-2 Your Lab Notebook

The lab notebook must

1. State what was done
2. State what was observed
3. Be understandable to someone else

The critical functions of your lab notebook are to state *what you did* and *what you observed*, and it should be *understandable by a stranger* who is trained in your discipline (chemistry in our case). The greatest error is writing ambiguous notes. After a few years, you may not be able to interpret your own notebook when memories of the experiment have faded. Writing in *complete sentences* is an excellent way to reduce this problem. Box 2-1 gives an example.

Without a doubt, somebody reading this book today is going to make an important discovery in the future and will seek a patent. The lab notebook is your legal record of your discovery. Therefore, each notebook page should be signed and dated. Anything of potential importance should also be signed and dated by a second person.

The measure of scientific "truth" is the ability to reproduce an experiment. A good lab notebook will allow you or anyone else to duplicate an experiment in the exact manner in which it was conducted the first time.

Beginning students find it useful (or required!) to write a complete description of an experiment, with sections describing the purpose, methods, results, and conclusions. Arranging your notebook to accept numerical data prior to coming to the lab is an excellent way to prepare for an experiment.

It is good practice to write a balanced chemical equation for every reaction that you use. This helps you understand what you are doing and may point out what you do not understand.

Do not rely on a computer for long-term storage of information. Even if a file survives, software or hardware required to read the file will become obsolete.

Record in your notebook the names of computer files where programs and data are stored. *Printed copies* of important data collected on a computer should be pasted into your notebook. The lifetime of a printed page is 10 to 100 times greater than that of a computer file.

Box 2-1 Dan's Lab Notebook Entry

Your lab notebook should (1) state what was done, (2) state what was observed, and (3) be understandable to someone else who is trained in your discipline. The passage below was extracted in 2002 from my notebook of 1974 when, as a "postdoc" at Albert Einstein College of Medicine, I began to isolate the iron storage protein ferritin. The complete procedure, in which protein was isolated and its purity assessed, occupied 3 weeks and 17 notebook pages. Phrases in brackets were added to help you understand the passage. I do not doubt that you can improve this description.

14 Sept 1974

Isolation of Human Spleen Ferritin

Based on R. R. Crichton et al., Biochem. J. 131, 51 (1973).

Procedure: Mince and homogenize spleen in ~4 vol H_2O
Heat to 70° for 5 min and cool on ice
Centrifuge at 3300×g for 20 min
Filter through filter paper
Precipitate with 50% $(NH_4)_2SO_4$ (= 313 g solid/L solution)
Centrifuge at 3300×g for 20 min
Dissolve in H_2O and di[redacted]

[redacted]

at that temperature with intermittent stirring for 5 min. It was then cooled in an ice bath to ~10° before centrifugation in the cold at 3300×g for 20 min (GSA head—4500 rpm). The red supernatant was filtered through Whatman #1 filter paper to give 218 mL solution, pH 6.4. The pH was raised to 7.5 with 10 M KOH and maintained between 7–8 during the addition of 68.2 g (50% saturation) $(NH_4)_2SO_4$. The solution [with precipitated protein] was left at RT [room temperature] overnight with 60 mg NaN_3 [a preservative]. Final pH = 7.6.

Later, there are tables of numerical data, graphs of results, and original, *well-labeled* instrument output pasted permanently into the notebook.

Ask Yourself

2-A. What are the three essential attributes of a lab notebook?

2-3 The Analytical Balance

Figure 2-2 shows a typical analytical **electronic balance** with a capacity of 100–200 g and a sensitivity of 0.01–0.1 mg. *Sensitivity* refers to the smallest increment of mass that can be measured. A *microbalance* weighs milligram quantities with a sensitivity of 0.1 μg. An electronic balance works by generating an electromagnetic force to

Balances are delicate and expensive. Be gentle when you place objects on the pan and when you adjust the knobs. A balance should be calibrated by measuring a set of standard weights at least every year.

Figure 2-2 Analytical electronic balance. Good-quality balances calibrate themselves with internal weights to correct for variations in the force of gravity, which can be as great as 0.3% from place to place. [Fisher Scientific, Pittsburgh, PA.]

balance the gravitational force acting on the object being weighed. The current required by the electromagnet is proportional to the mass being weighed.

Figure 2-3 shows the principle of operation of a single-pan **mechanical balance.** The balance beam is suspended on a sharp *knife edge*. The mass of the pan hanging from the balance point (another knife edge) at the left is balanced by a counterweight at the right. You place the object to be weighed on the pan and adjust knobs that remove standard weights from the bar above the pan. The balance beam

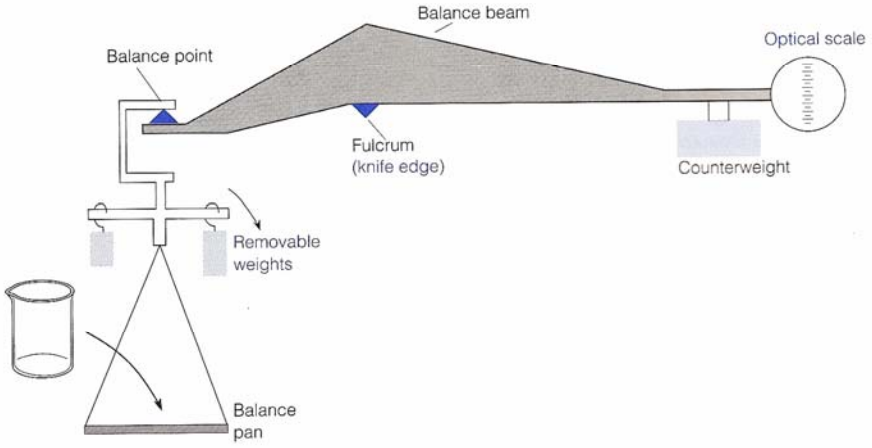


Figure 2-3 Single-pan mechanical balance. After placing an object on the pan, we detach removable weights until the balance beam is restored as close as possible to its original position. The remaining small difference is read on the optical scale.

is restored close to its original position when the weights removed from the bar are nearly equal to the mass on the pan. The slight difference from the original position is shown on an optical scale, whose reading is added to that of the knobs.

A mechanical balance should be in its arrested position when you load or unload the pan and in the half-arrested position when you are dialing weights. This practice prevents abrupt forces that would wear down the knife edges and decrease the sensitivity of the balance.

Using a Balance

To weigh a chemical, place a clean receiving vessel on the balance pan. The mass of the empty vessel is called the **tare**. On most electronic balances, the tare can be set to 0 by pressing a button. Add chemical to the vessel and read the new mass. If there is no automatic tare operation, the mass of the empty vessel should be subtracted from that of the filled vessel. Do not place chemicals directly on the weighing pan. This precaution protects the balance from corrosion and allows you to recover all the chemical being weighed.

An alternate procedure, called "weighing by difference," is necessary for **hygroscopic** reagents, which rapidly absorb moisture from the air. First weigh a capped bottle containing dry reagent. Then quickly pour some reagent from the weighing bottle into a receiver. Cap the weighing bottle and weigh it again. The difference is the mass of reagent delivered. With an electronic balance, set the initial mass of the weighing bottle to 0 with the tare button. Then deliver reagent from the bottle and reweigh the bottle. The negative reading on the balance is the mass of reagent delivered from the bottle.[3]

Clean up spills on the balance and do not allow chemicals to get into the mechanism below the pan. Use a paper towel or tissue to handle the vessel that you are weighing, because fingerprints will change its mass. Samples should be at *ambient temperature* (the temperature of the surroundings) when weighed to prevent errors due to convective air currents. The doors of the balance in Figure 2-2 must be closed during weighing so that air currents do not disturb the pan. A top-loading balance without sliding doors has a fence around the pan to deflect air currents. Sensitive balances should be placed on a heavy table, such as a marble slab, to minimize the effect of vibrations on the reading. Use the bubble meter and adjustable feet of a balance to keep it level.

Buoyancy

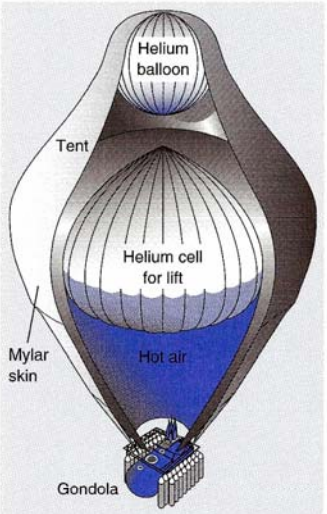
When you swim, your weight in the water is nearly zero, which is why people can float. **Buoyancy** is the upward force exerted on an object in a liquid or gaseous fluid. An object weighed in air appears lighter than its actual mass by an amount equal to the mass of air that it displaces. The true mass is the mass measured in vacuum. The standard weights in a balance also are affected by buoyancy, so they weigh less in air than they would in vacuum. A buoyancy error occurs whenever the density of the object being weighed is not equal to the density of the standard weights.

If mass m' is read from a balance, the true mass m is

Buoyancy equation:

$$m = \frac{m'\left(1 - \frac{d_a}{d_w}\right)}{\left(1 - \frac{d_a}{d}\right)} \qquad (2\text{-}1)$$

where d_a is the density of air (0.001 2 g/mL near 1 bar and 25°C); d_w is the density of balance weights (8.0 g/mL); and d is the density of the object being weighed.



The *Breitling Orbiter 3* in 1999 was the first balloon to fly around the world. Previous balloons could not carry enough propane fuel for such a trip. The design of the *Breitling Orbiter 3* keeps the temperature of the large, inner helium cell as constant as possible, so the buoyancy variation between the warm day and the cold night is minimal. During the day, the sun heats the helium cell, which expands, thereby increasing its buoyancy and its ability to keep the vessel aloft. If the temperature rises too much, a solar-powered fan brings in cool air to keep the vessel from rising to an undesired altitude. At night, the helium cell cools and shrinks, which reduces its buoyancy. To keep the balloon aloft at night, heat from burning propane is required. The double-wall design reduces radiational cooling of the helium cell and decreases the requirement for propane.

Example **Buoyancy Correction**

Find the true mass of water (density = 1.00 g/mL) if the apparent mass is 100.00 g.

SOLUTION Equation 2-1 gives the true mass:

$$m = \frac{100.00\ \text{g}\left(1 - \dfrac{0.001\ 2\ \text{g/mL}}{8.0\ \text{g/mL}}\right)}{\left(1 - \dfrac{0.001\ 2\ \text{g/mL}}{1.00\ \text{g/mL}}\right)} = 100.11\ \text{g}$$

Test Yourself Find the true mass of 28.0 wt% ammonia (density = 0.90 g/mL) when the apparent mass is 20.000 g. (**Answer:** 20.024 g)

The buoyancy error for water is 0.11%, which is significant for many purposes. For solid NaCl with a density of 2.16 g/mL, the error is 0.04%.

Ask Yourself

2-B. (a) Buoyancy corrections are most critical when you calibrate glassware such as a volumetric flask to see how much volume it actually holds. Suppose that you fill a 25-mL volumetric flask with distilled water and find that the mass of water in the flask measured in air is 24.913 g. What is the true mass of the water?
(b) You made the measurement when the lab temperature was 21°C, at which the density of water is 0.998 00 g/mL. What is the true volume of water contained in the volumetric flask?

2-4 Burets

A **buret**[4] is a precisely manufactured glass tube with graduations enabling you to measure the volume of liquid delivered through the *stopcock* (the valve) at the bottom (Figure 2-4*a*). The numbers on the buret increase from top to bottom (with 0 mL near the top). A volume measurement is made by reading the level before and after draining liquid from the buret and subtracting the first reading from the second reading. The graduations of Class A burets (the most accurate grade) are certified to meet the tolerances in Table 2-1. For example, if the reading of a 50-mL buret is 32.50 mL, the true volume can be anywhere in the range 32.45 to 32.55 mL and still be within the manufacturer's stated tolerance of ±0.05 mL.

Table 2-1 **Tolerances of Class A burets**

Buret volume (mL)	Smallest graduation (mL)	Tolerance (mL)
5	0.01	±0.01
10	0.05 *or* 0.02	±0.02
25	0.1	±0.03
50	0.1	±0.05
100	0.2	±0.10

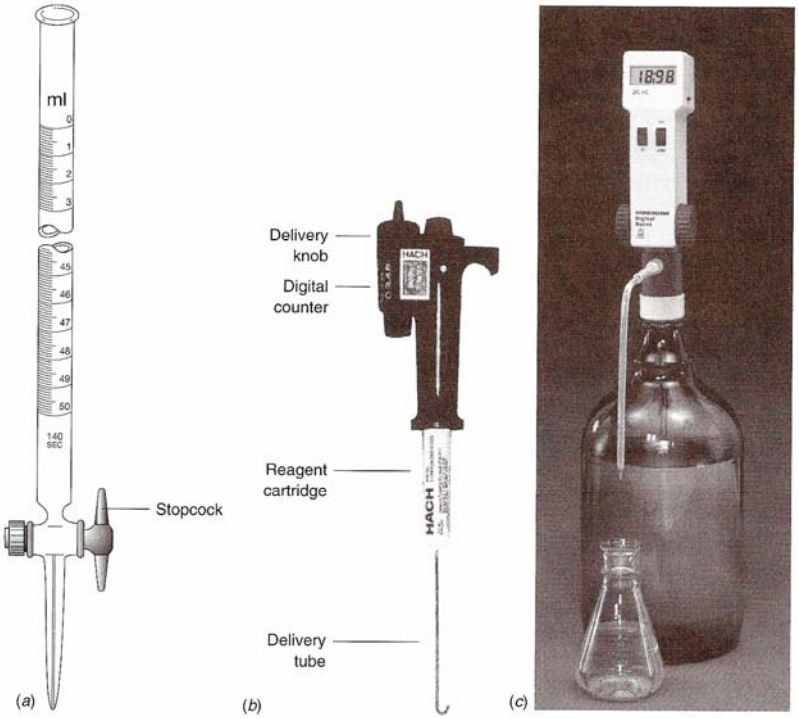


Figure 2-4 (*a*) Glass buret with Teflon stopcock. Cover your buret with a loose-fitting cap to keep dust out and reduce evaporation. [Fisher Scientific, Pittsburgh, PA.] (*b*) Digital titrator with its plastic cartridge containing reagent carries out the same function as a buret for analyses in the field. [Hach Co., Loveland, CO.] (*c*) Battery-operated electronic buret with digital readout delivers 0.01-mL increments from a reagent bottle. [Cole-Parmer Co., Niles, IL.]

When reading the liquid level in a buret, your eye should be at the same height as the top of the liquid. If your eye is too high, the liquid seems to be higher than it actually is. If your eye is too low, the liquid appears too low. The error that occurs when your eye is not at the same height as the liquid is called **parallax error**.

The **meniscus** is the curved upper surface of liquid in the glass buret in Figure 2-5. Water has a concave meniscus because water is attracted to glass and climbs slightly up the glass. It is helpful to use black tape on a white card as a background for locating the meniscus. Align the top of the tape with the bottom of the meniscus and read the position on the buret. Highly colored solutions may appear to have a double meniscus, either of which you can use. Volume is determined by subtracting one reading from another, so the important point is to read the position of the meniscus reproducibly. Always estimate the reading to the nearest tenth of the division between marks.

The thickness of a graduation line on a 50-mL buret corresponds to about 0.02 mL. To use the buret most accurately, consider the *top* of a graduation line to be 0. When the meniscus is at the bottom of the same graduation line, the reading is 0.02 mL greater.

A drop from a 50-mL buret is about 0.05 mL. Near the end point of a titration, try to deliver less than one drop at a time so that you can locate the end point more precisely than ± 0.05 mL. To deliver a fraction of a drop, carefully open the stopcock until part of a drop is hanging from the buret tip. Then touch the inside wall of the receiving flask to the buret tip to transfer the droplet to the flask. Carefully tip the flask

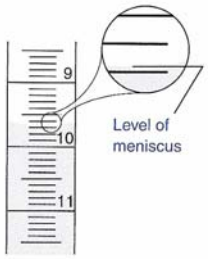
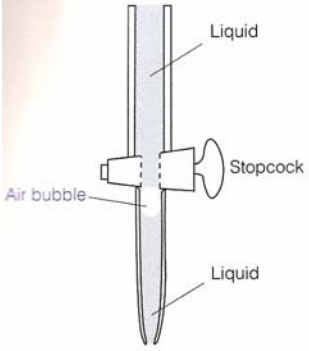


Figure 2-5 Buret with the meniscus at 9.68 mL. Estimate the reading of any scale to the nearest tenth of a division. Because this buret has 0.1-mL divisions, we estimate the reading to 0.01 mL.



igure 2-6 Air bubble beneath ıe stopcock should be expelled efore you use a buret.

perating a buret:

read bottom of concave meniscus
estimate reading to 1/10 of a division
avoid parallax
account for graduation thickness in readings
drain liquid slowly
wash buret with new solution
deliver fraction of a drop near end point
eliminate air bubble before use

so that the main body of liquid washes over the newly added droplet. Then swirl the flask to mix the contents. Near the end of a titration, tip and rotate the flask often to ensure that droplets on the wall containing unreacted analyte contact the bulk solution.

Liquid should drain evenly down the wall of a buret. The tendency of liquid to stick to the glass is reduced by draining the buret slowly (<20 mL/min). If many droplets stick to the wall, clean the buret with detergent and a buret brush. If this cleaning is insufficient, soak the buret in peroxydisulfate–sulfuric acid cleaning solution prepared by your instructor.[5] Cleaning solution eats clothing and people, as well as grease in the buret. Volumetric glassware should not be soaked in alkaline solutions, which attack glass. (A 5 wt% NaOH solution at 95°C dissolves Pyrex glass at a rate of 9 μm/h.)

A common buret error is caused by failure to expel the air bubble often found beneath the stopcock (Figure 2-6). A bubble present at the start of the titration may be filled with liquid during the titration. Therefore some volume that drained out of the graduated part of the buret did not reach the titration vessel. Usually the bubble can be dislodged by draining the buret for a second or two with the stopcock wide open. A tenacious bubble can be expelled by carefully shaking the buret while draining it into a sink.

Before you fill a buret with fresh solution, it is a wonderful idea to rinse the buret several times with small portions of the new solution, discarding each wash. It is not necessary to fill the entire buret with wash solution. Simply tilt the buret so that its whole surface contacts the wash liquid. This same technique should be used with any vessel (such as a spectrophotometer cuvet or a pipet) that is reused without drying.

The *digital titrator* in Figure 2-4*b* is useful for conducting titrations in the field where samples are collected. The counter tells how much reagent from the cartridge has been dispensed by rotation of the delivery knob. Its accuracy of 1% is 10 times poorer than the accuracy of a glass buret, but many measurements do not require higher accuracy. The battery-operated *electronic buret* in Figure 2-4*c* fits on a reagent bottle and delivers up to 99.99 mL in 0.01-mL increments displayed on a digital readout.

Microscale Titrations (A "Green" Idea)

"Microscale" experiments decrease costs, consumption of reagents, and generation of waste. A student buret can be constructed from a 2-mL pipet graduated in 0.01-mL intervals.[6] Volume can be read to 0.001 mL, and titrations can be carried out with a precision of 1%.

2-5 Volumetric Flasks

A **volumetric flask** (Figure 2-7, Table 2-2) is calibrated to contain a particular volume of solution at 20°C when the bottom of the meniscus is adjusted to the center of the mark on the neck of the flask (Figure 2-8). Most flasks bear the label "TC 20°C," which means *to contain* at 20°C. (Other types of glassware may be calibrated *to deliver,* "TD," their indicated volume.) The temperature of the container is relevant because liquid and glass expand when heated.

We use a volumetric flask to prepare a solution of known volume. Typically, reagent is weighed into the flask, dissolved, and diluted to the mark. The mass of reagent and final volume are therefore known. Dissolve the reagent in the flask in *less* than the final volume of liquid. Add more liquid and mix the solution again. Make the final volume adjustment with as much well-mixed liquid in the flask as possible. (When two different liquids are mixed, there is generally a small volume change. The total volume is *not* the sum of the two volumes that were mixed. By swirling the liquid in a nearly full volumetric flask before the liquid reaches the thin neck, you

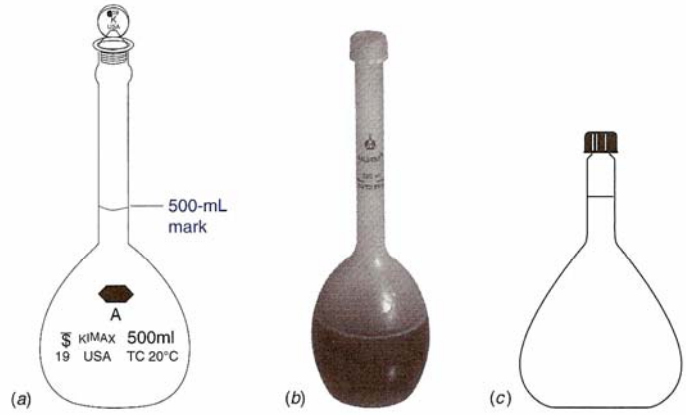


Figure 2-7 (*a*) Class A glass volumetric flask meets tolerances in Table 2-2. [A. H. Thomas Co., Philadelphia, PA.] (*b*) Class B polypropylene plastic flask for trace analysis (ppb concentrations) in which analyte might be lost by adsorption (sticking) on glass or contaminated with previously adsorbed species. A plastic flask is also required for reagents such as HF or hot, basic solutions that react with glass. Class B flasks are less accurate than Class A flasks, with twice the tolerances in Table 2-2. [Fisher Scientific, Pittsburgh, PA.] (*c*) Short-form volumetric flask with Teflon-lined screw cap fits on analytical balance. Teflon prevents solutions from attacking the inside of the cap.

minimize the change in volume when the last liquid is added.) For best control, add the final drops of liquid with a pipet, *not a squirt bottle*. After adjusting the liquid to the correct level, hold the cap firmly in place and invert the flask several times to complete mixing. Before the liquid is homogeneous, we observe streaks (called *schlieren*) arising from regions that refract light differently. After the schlieren are gone, invert the flask a few more times to ensure complete mixing.

Glass is notorious for *adsorbing* traces of chemicals—especially cations. **Adsorption** means to stick to the surface. (In contrast, **absorption** means to take inside, as a sponge takes up water.) For critical work, **acid wash** the glassware to replace low concentrations of cations on the glass surface with H^+. To do this, soak already thoroughly cleaned glassware in 3–6 M HCl or HNO_3 (in a fume hood) for >1 h, followed by several rinses with distilled water and a final soak in distilled water. The HCl can be reused many times, as long as it is only used for soaking clean glassware.

As an example, high purity nitric acid was delivered from a glass pipet that had been washed normally without acid and another that had been acid washed. The level of the transition elements Ti, Cr, Mn, Fe, Co, Ni, Cu, and Zn in acid delivered from the acid-washed pipet was below the detection level of 0.01 ppb (0.01 ng/g). The concentration of each transition element in acid delivered from the pipet that had not been acid washed was in the range 0.5 to 9 ppb.[7]

Adsorption: to bind a substance on the surface

Absorption: to bind a substance internally

Ask Yourself

2-C. How would you use a volumetric flask to prepare 250.0 mL of 0.150 0 M K_2SO_4?

Table 2-2 Tolerances of Class A volumetric flasks

Flask capacity (mL)	Tolerance (mL)	Flask capacity (mL)	Tolerance (mL)
1	±0.02	100	±0.08
2	±0.02	200	±0.10
5	±0.02	250	±0.12
10	±0.02	500	±0.20
25	±0.03	1 000	±0.30
50	±0.05	2 000	±0.50

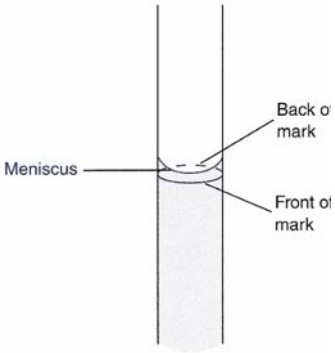


Figure 2-8 Proper position of the meniscus: at the center of the ellipse formed by the front and back of the calibration mark when viewed from above or below the level of the mark. Volumetric flasks and transfer pipets are calibrated to this position.

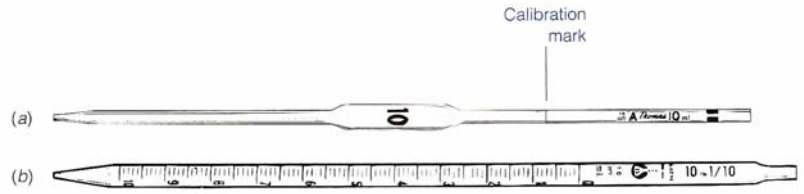


Figure 2-9 (*a*) Transfer pipet. Do not blow out the last drop. (*b*) Measuring (Mohr) pipet. [A. H. Thomas Co., Philadelphia, PA.]

2-6 Pipets and Syringes

Pipets deliver known volumes of liquid. The *transfer pipet* in Figure 2-9 is calibrated to deliver one fixed volume. The last drop of liquid does not drain out of the pipet; *it should not be blown out*. The *measuring pipet* is calibrated to deliver a variable volume, which is the difference between the initial and final volumes. A measuring pipet could be used to deliver 5.6 mL by starting delivery at the 1-mL mark and terminating at the 6.6-mL mark.

Do not blow the last drop out of a transfer pipet.

A transfer pipet is more accurate than a measuring pipet. Tolerances for Class A (the most accurate grade) transfer pipets in Table 2-3 are the allowed error in the volume that is actually delivered.

Table 2-3 Tolerances of Class A transfer pipets

Volume (mL)	Tolerance (mL)
0.5	±0.006
1	±0.006
2	±0.006
3	±0.01
4	±0.01
5	±0.01
10	±0.02
15	±0.03
20	±0.03
25	±0.03
50	±0.05
100	±0.08

Using a Transfer Pipet

Using a rubber bulb, *not your mouth*, suck liquid up past the calibration mark. It is a good idea to discard one or two pipet volumes of liquid to remove traces of previous reagents from the pipet. After taking up a third volume past the calibration mark, quickly replace the bulb with your index finger at the end of the pipet. The liquid should still be above the mark after this maneuver. Pressing the pipet against the bottom of the vessel while removing the rubber bulb helps prevent liquid from draining while you put your finger in place. Wipe the excess liquid off the outside of the pipet with a clean tissue. *Touch the tip of the pipet to the side of a beaker* and drain the liquid until the bottom of the meniscus just reaches the center of the mark, as in Figure 2-8. Touching the beaker wall draws liquid from the pipet without leaving part of a drop hanging from the pipet when the level reaches the calibration mark.

Transfer the pipet to the desired receiving vessel and drain it *while holding the tip against the wall of the vessel*. After the pipet stops draining, hold it against the wall for a few more seconds to complete draining. *Do not blow out the last drop*. The pipet should be nearly vertical at the end of delivery. When you finish with a pipet, it should be rinsed with distilled water or soaked in a pipet container until it is cleaned. Solutions should never be allowed to dry inside a pipet because dry residue is difficult to remove.

Accuracy: difference between delivered volume and desired volume
Precision: reproducibility of replicate deliveries

Micropipets

A micropipet (Figure 2-10) is used to deliver volumes of 1 to 1 000 μL (1 μL = 10^{-6} L) with accuracies given in Table 2-4. The liquid is contained in the disposable plastic tip. Micropipets may have a metal barrel on the inside that can be corroded by pipetting

Table 2-4 Accuracy (%) of micropipets

Pipet volume (μL)	At 10% of pipet volume	At 100% of pipet volume	Pipet volume (μL)	Accuracy (%)
	Adjustable volume		*Fixed volume*	
2	±8	±1.2	10	±0.8
10	±2.5	±0.8	25	±0.8
25	±4.5	±0.8	100	±0.5
100	±1.8	±0.6	500	±0.4
300	±1.2	±0.4	1 000	±0.3
1 000	±1.6	±0.3		

Precision is typically 2–3 times smaller (better) than the accuracy.
SOURCE: Hamilton Co., Reno, NV.

volatile acids such as concentrated HCl. Corrosion slowly diminishes the accuracy of the pipet.

To use a micropipet, place a fresh tip tightly on the barrel. Tips are contained in a package or dispenser so that you do not handle (and contaminate) the points with your fingers. Set the desired volume with the knob at the top of the pipet. Depress the plunger to the first stop, which corresponds to the selected volume. Hold the pipet *vertically*, dip it 3–5 mm into the reagent solution, and *slowly* release the plunger to suck up liquid. Withdraw the tip from the liquid by sliding it along the wall of the vessel to remove liquid from the outside of the tip. To dispense liquid, touch the micropipet tip to the wall of the receiver and gently depress the plunger to the first stop. After a few seconds to allow liquid to drain down the wall of the pipet tip, depress the plunger farther to squirt out the last liquid. It is a good idea to clean and wet a fresh tip by taking up and discarding two or three squirts of reagent first. The tip can be discarded or rinsed well with a squirt bottle and reused. When you use a squirt bottle, never touch the tip of the squirt bottle to anything, to avoid contaminating the squirt bottle.

The volume of liquid taken into the tip depends on the angle at which the pipet is held and how far beneath the surface of reagent the tip is held during uptake. As internal parts wear out, both precision and accuracy can decline by an *order of magnitude* (a factor of 10). Micropipets require periodic cleaning, seal replacement, and lubrication. You can check performance by weighing the amount of water delivered from a micropipet. Monthly calibration to identify pipets in need of repair is recommended.

A microliter *syringe*, such as that in Figure 2-11, dispenses volumes in the range 1 to 500 μL with accuracy and precision near 1%. When using a syringe, take up and discard several volumes of liquid to wash the glass and remove air bubbles from the barrel. The steel needle is attacked by strong acid and will contaminate strongly acidic solutions with iron.

Figure 2-11 Hamilton syringe with a volume of 1 μL and graduations of 0.01 μL on the glass barrel. [Hamilton Co., Reno, NV.]

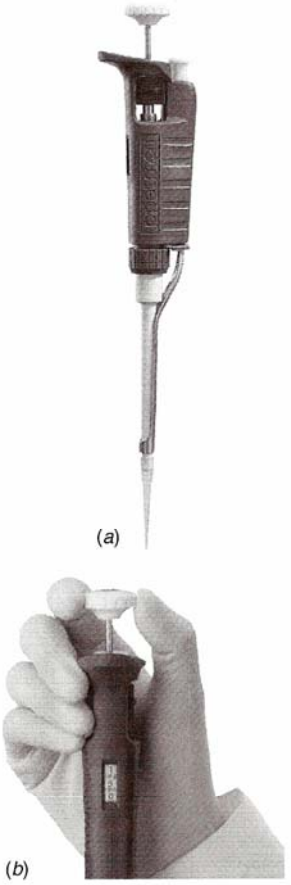


Figure 2-10 (*a*) Micropipet with disposable plastic tip. (*b*) Volume selection dial set to 150 μL. [Rainin Instrument Co., Emeryville, CA.]

When you use a squirt bottle, *never* touch the tip to anything.

Ask Yourself

2-D. Which is more accurate, a transfer pipet or a measuring pipet? How much is the uncertainty in microliters when you deliver **(a)** 10 μL or **(b)** 100 μL from a 100-μL adjustable micropipet?

2-7 Filtration

In **gravimetric analysis**, the mass of product from a reaction is measured to determine how much unknown was present. Precipitates from gravimetric analyses are collected by filtration, washed, and then dried. Most precipitates are collected in a *fritted-glass funnel* with suction to speed filtration (Figure 2-12). The porous glass plate in the funnel allows liquid to pass but retains solids. Filters with coarse, medium, and fine pores are available to collect precipitates with large, medium, or small particle size. The finer the filter, the slower the filtration. The empty crucible is first dried at 110°C and weighed. After collecting solid and drying again, the crucible and its contents are weighed a second time to determine the mass of solid.

Liquid from which a substance precipitates or crystallizes is called the **mother liquor**. Liquid that passes through the filter is called **filtrate**.

In some gravimetric procedures, **ignition** (heating at high temperature over a burner or in a furnace) is used to convert a precipitate to a known, constant composition. For example, Fe^{3+} precipitates as hydrated $Fe(OH)_3$ with variable composition. Ignition converts it to Fe_2O_3 prior to weighing. When a gravimetric precipitate is to be ignited, it is collected in **ashless filter paper**, which leaves little residue when burned.

To use filter paper with a conical glass funnel, fold the paper into quarters, tear off one corner (to allow a firm fit into the funnel), and place the paper in the funnel (Figure 2-13). The filter paper should fit snugly and be seated with some distilled water. When liquid is poured in, an unbroken stream of liquid should fill the stem of the funnel. The weight of liquid in the stem helps speed filtration.

For filtration, pour the slurry of precipitate in the mother liquor down a glass rod to prevent splattering (Figure 2-14). (A *slurry* is a suspension of solid in liquid.) Dislodge any particles adhering to the beaker or rod with a **rubber policeman**, which is a flattened

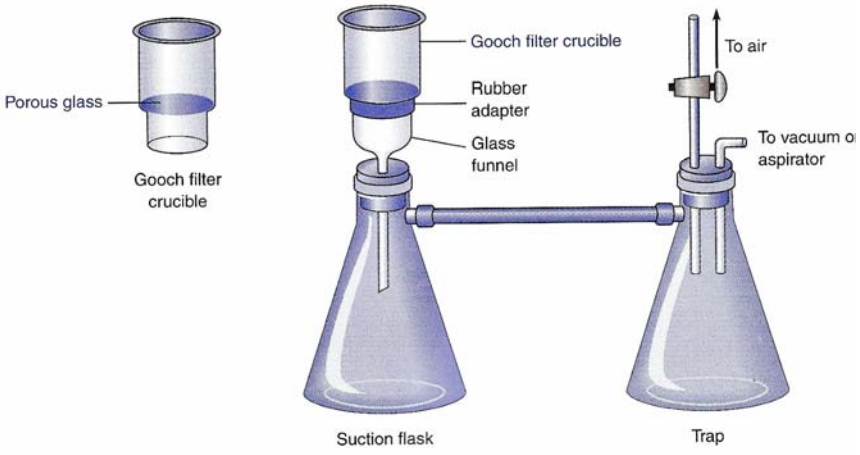


Figure 2-12 Filtration with a Gooch filter crucible that has a porous (*fritted*) glass disk through which liquid can pass. Suction is provided by a vacuum line at the lab bench or by an *aspirator* that uses flowing water from a tap to create a vacuum. The trap prevents backup of filtrate into the vacuum system or backup of water from the aspirator into the suction flask.

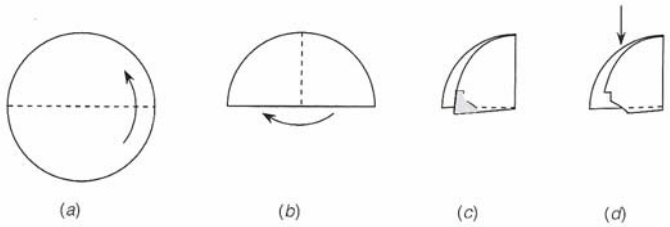


Figure 2-13 Folding filter paper for a conical funnel. (*a*) Fold the paper in half and (*b*) in half again. (*c*) Tear off a corner to better seat the paper in the funnel. (*d*) Open the side that was not torn and fit the paper in the funnel.

piece of rubber at the end of a glass rod. Use a jet of appropriate wash liquid from a squirt bottle to transfer particles from the rubber and glassware to the filter. If the precipitate is going to be ignited, particles remaining in the beaker should be wiped onto a small piece of moist filter paper, which is then added to the filter to be ignited.

2-8 Drying

Reagents, precipitates, and glassware are usually dried in an oven at 110°C. (Some chemicals require other temperatures.) Label everything that you put in the oven. Use a beaker and watchglass (Figure 2-15) to minimize contamination by dust during drying. It is good practice to cover all vessels on the benchtop to prevent dust contamination.

We measure the mass of a gravimetric precipitate by weighing a dry, empty filter crucible before the procedure and weighing the same crucible containing dry product after the procedure. To weigh the empty crucible, first bring it to "constant mass" by drying in the oven for 1 h or longer and then cooling for 30 min in a *desiccator*. Weigh the crucible and then heat it again for about 30 min. Cool it and reweigh it. When successive weighings agree to ±0.3 mg, the filter has reached "constant mass." A microwave oven can be used instead of an electric oven for drying reagents, precipitates, and crucibles. Try an initial heating time of 4 min, with subsequent 2-min heatings.

A **desiccator** (Figure 2-16) is a closed chamber containing a drying agent called a **desiccant**. The lid is greased to make an airtight seal. Desiccant is placed in the

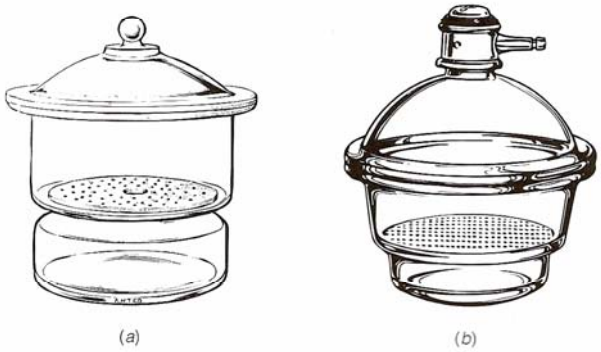


Figure 2-16 (*a*) Ordinary desiccator. (*b*) Vacuum desiccator, which can be evacuated through the side arm and then sealed by rotating the joint containing the side arm. Drying is more efficient at low pressure. Drying agents (*desiccants*) are placed at the bottom of each desiccator below the porous porcelain plate. [A. H. Thomas Co., Philadelphia, PA.]

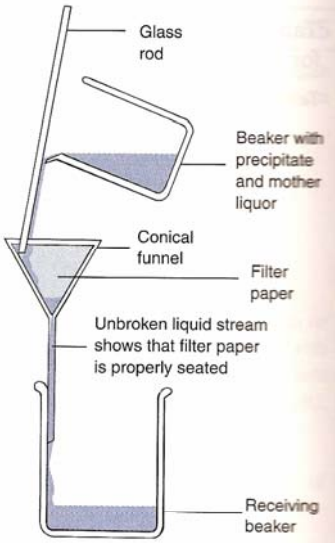


Figure 2-14 Filtering a precipitate.

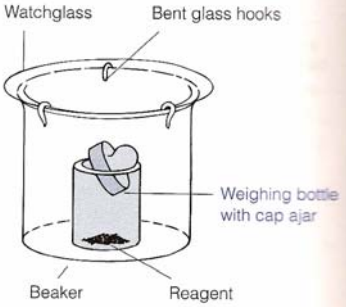


Figure 2-15 Use a watchglass as a dust cover while drying reagents or crucibles in the oven.

Table 2-5 Correction factors for volumetric calibration

Temperature (°C)	Correction factor (mL/g)[a]
15	1.002 0
16	1.002 1
17	1.002 3
18	1.002 5
19	1.002 7
20	1.002 9
21	1.003 1
22	1.003 3
23	1.003 5
24	1.003 8
25	1.004 0
26	1.004 3
27	1.004 6
28	1.004 8
29	1.005 1
30	1.005 4

a. Factors are based on the density of water and are corrected for buoyancy with Equation 2-1.

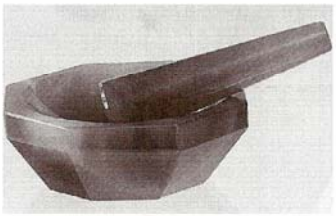


Figure 2-17 Agate mortar and pestle. The mortar is the base and the pestle is the grinding tool. Agate is very hard and expensive. Less expensive porcelain mortars are widely used, but they are somewhat porous and easily scratched. These properties can lead to contamination of the sample by porcelain particles or by traces of previous samples embedded in the porcelain. [Thomas Scientific, Swedesboro, NJ.]

bottom beneath the perforated disk. Common desiccants in approximate order of decreasing efficiency are magnesium perchlorate ($Mg(ClO_4)_2$) > barium oxide (BaO) ≈ alumina (Al_2O_3) ≈ phosphorus pentoxide (P_4O_{10}) >> calcium chloride ($CaCl_2$) ≈ calcium sulfate ($CaSO_4$, called Drierite) ≈ silica gel (SiO_2). After placing a hot object in the desiccator, leave the lid cracked open for a minute until the object has cooled slightly. This practice prevents the lid from popping open when the air inside warms up. To open a desiccator, slide the lid sideways rather than trying to pull it straight up.

2-9 Calibration of Volumetric Glassware

Volumetric glassware can be calibrated to measure the volume that is actually contained in or delivered by a particular piece of equipment. Calibration is done by measuring the mass of water contained or delivered and using Table 2-5 to convert mass to volume:

$$\text{true volume} = (\text{mass of water}) \times (\text{correction factor in Table 2-5}) \qquad (2\text{-}2)$$

To calibrate a 25-mL transfer pipet, first weigh an empty weighing bottle like the one in Figure 2-15. Then fill the pipet to the mark with distilled water, drain it into the weighing bottle, and put on the lid to prevent evaporation. Weigh the bottle again to find the mass of water delivered from the pipet. Use Equation 2-2 to convert mass to volume.

Example **Calibration of a Pipet**

An empty weighing bottle had a mass of 10.283 g. After water was added from a 25-mL pipet, the mass was 35.225 g. The temperature was 23°C. Find the volume of water delivered by the pipet.

SOLUTION The mass of water is 35.225 − 10.283 = 24.942 g. From Equation 2-2 and Table 2-5, the volume of water is (24.942 g)(1.003 5 mL/g) = 25.029 mL.

Test Yourself If the temperature had been 29°C, and the mass of water was 24.942 g, what volume was delivered? (**Answer:** 25.069 mL)

Ask Yourself

2-E. A 10-mL pipet delivered 10.000 0 g of water at 15°C to a weighing bottle. What is the true volume of the pipet?

2-10 Methods of Sample Preparation

In the analytical process chart in Box 0-1, a homogeneous laboratory sample must be prepared from a representative bulk sample. You can homogenize solids by grinding them to fine powder with a **mortar and pestle** (Figure 2-17) or by dissolving the entire sample.

Dissolving Inorganic Materials with Strong Acids

The acids HCl, HBr, HF, H_3PO_4, and dilute H_2SO_4 dissolve most metals (M) by the reaction

$$M(s) + nH^+(aq) \xrightarrow{\text{heat}} M^{n+}(aq) + \frac{n}{2}H_2(g)$$

Many other inorganic substances also can be dissolved. Some anions react with H^+ to form **volatile** products (species that evaporate easily), which are lost from hot solutions in open vessels. Examples include carbonate ($CO_3^{2-} + 2H^+ \longrightarrow H_2CO_3 \longrightarrow CO_2\uparrow + H_2O$) and sulfide ($S^{2-} + 2H^+ \longrightarrow H_2S\uparrow$). Hot hydrofluoric acid dissolves silicates found in most rocks. HF also attacks glass, so it is used in Teflon, polyethylene, silver, or platinum vessels. Teflon is inert to attack by most chemicals and can be used up to 260°C.

Substances that do not dissolve in the acids above may dissolve as a result of oxidation by HNO_3 or concentrated H_2SO_4. Nitric acid attacks most metals, but not Au and Pt, which dissolve in the 3:1 (vol:vol) mixture of HCl:HNO_3 called *aqua regia.*

Acid dissolution is conveniently carried out with a Teflon-lined **bomb** (a sealed vessel, Figure 2-18) in a microwave oven, which heats the contents to 200°C in a minute. The bomb cannot be made of metal, which absorbs microwaves. The bomb should be cooled prior to opening to prevent loss of volatile products.

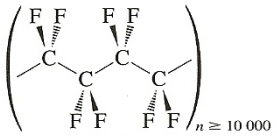
HCl	hydrochloric acid
HBr	hydrobromic acid
HF	hydrofluoric acid
H_3PO_4	phosphoric acid
H_2SO_4	sulfuric acid
HNO_3	nitric acid

HF is extremely harmful to touch or breathe. Flood the affected area with water, coat the skin with calcium gluconate (or another calcium salt), and seek medical help.

Fusion

Inorganic substances that do not dissolve in acid can usually be dissolved by a hot, molten inorganic **flux**, examples of which are lithium tetraborate ($Li_2B_4O_7$) and sodium hydroxide (NaOH). Mix the finely powdered unknown with 2 to 20 times its mass of solid flux, and **fuse** (melt) the mixture in a platinum–gold alloy crucible at

Teflon is a *polymer* (a chain of repeating units) with the structure



Teflon

Carbon atoms are in the plane of the page. A solid wedge is a bond coming out of the page toward you and a dashed wedge is a bond going behind the page.

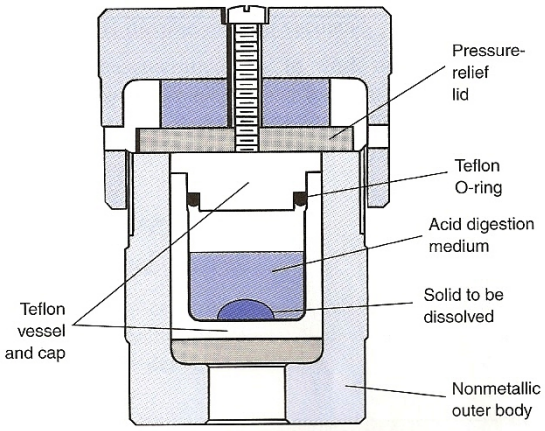


Figure 2-18 Microwave digestion bomb lined with Teflon. A typical 23-mL vessel can be used to digest as much as 1 g of inorganic material (or 0.1 g of organic material, which releases a great deal of gaseous CO_2) in as much as 15 mL of concentrated acid. The outer container maintains its strength up to 150°C, but rarely rises above 50°C. If the internal pressure exceeds 80 bar, the cap deforms and releases the excess pressure. [Parr Instrument Co., Moline, IL.]

300° to 1 200°C in a furnace or over a burner. When the sample is homogeneous, carefully pour the molten flux into a beaker containing 10 wt% aqueous HNO_3 to dissolve the product.

Digestion of Organic Substances

To analyze N, P, halogens (F, Cl, Br, I), and metals in an organic compound, first decompose the compound by combustion (described in Section 7-4) or by *digestion*. In **digestion**, a substance is decomposed and dissolved by a reactive liquid. For this purpose, add sulfuric acid or a mixture of H_2SO_4 and HNO_3 to an organic substance and gently boil (or heat it in a microwave bomb) for 10 to 20 min until all particles have dissolved and the solution has a uniform black appearance. After cooling, destroy the dark color by adding hydrogen peroxide (H_2O_2) or HNO_3, and heat again. Analyze the decomposed sample after digestion.

Extraction

In **extraction**, analyte is dissolved in a solvent that does not dissolve the entire sample and does not decompose the analyte. In a typical extraction of pesticides from soil, a mixture of soil plus the solvents acetone and hexane is placed in a Teflon-lined bomb and heated by microwaves to 150°C. This temperature is 50° to 100° higher than the boiling points of the individual solvents in an open vessel at atmospheric pressure. Soluble pesticides dissolve, but most of the soil remains behind. To complete the analysis, analyze the solution by chromatography, which is described in Chapters 21 through 23.

Ask Yourself

2-F. Lead sulfide (PbS) is a black solid that is sparingly soluble in water but dissolves in concentrated HCl. If such a solution is boiled to dryness, white, crystalline lead chloride ($PbCl_2$) remains. What happened to the sulfide?

Key Equation

Buoyancy

$$m = m' \left(1 - \frac{d_a}{d_w}\right) \Big/ \left(1 - \frac{d_a}{d}\right)$$

m = true mass; m' = mass measured in air

d_a = density of air (0.001 2 g/mL near 1 bar and 25°C)

d_w = density of balance weights (8.0 g/mL)

d = density of object being weighed

Important Terms

absorption
acid wash
adsorption
ashless filter paper
bomb
buoyancy
buret
desiccant
desiccator
digestion
electronic balance
extraction
filtrate
flux
fusion

gravimetric analysis
green chemistry
hygroscopic
ignition
mechanical balance
meniscus
mortar and pestle
mother liquor
parallax error
pipet
rubber policeman
tare
volatile
volumetric flask

Problems

2-1. What do the symbols TD and TC mean on volumetric glassware?

2-2. When would it be preferable to use a plastic volumetric flask instead of a glass flask?

2-3. What is the purpose of the trap in Figure 2-12? What does the watchglass do in Figure 2-15?

2-4. Distinguish absorption from adsorption. When you heat glassware in a drying oven, are you removing absorbed or adsorbed water?

2-5. What is the difference between digestion and extraction?

2-6. What is the true mass of water if the mass measured in air is 5.397 4 g?

2-7. Pentane (C_5H_{12}) is a liquid with a density of 0.626 g/mL. Find the true mass of pentane when the mass weighed in air is 14.82 g.

2-8. Ferric oxide (Fe_2O_3, density = 5.24 g/mL) obtained from ignition of a gravimetric precipitate weighed 0.296 1 g in the atmosphere. What is the true mass in vacuum?

2-9. Your professor has recruited you to work in her lab to help her win the Nobel Prize. It is critical that your work be as accurate as possible. Rather than using the stated volumes of glassware in the lab, you decide to calibrate each piece. An empty 10-mL volumetric flask weighed 10.263 4 g. When filled to the mark with distilled water at 20°C, it weighed 20.214 4 g. What is the true volume of the flask?

2-10. Water from a 5-mL pipet was drained into a weighing bottle whose empty mass was 9.974 g to give a new mass of 14.974 g at 26°C. Find the volume of the pipet.

2-11. Water was drained from a buret between the 0.12- and 15.78-mL marks. The apparent volume was 15.78 − 0.12 = 15.66 mL. Measured in air at 25°C, the mass of water delivered was 15.569 g. What was the true volume?

2-12. Glass is a notorious source of metal ion contamination. Three glass bottles were crushed and sieved to collect 1-mm pieces.[8] To see how much Al^{3+} could be extracted, 200 mL of a 0.05 M solution of the metal-binding compound EDTA was stirred with 0.50 g of ~1-mm glass particles in a polyethylene flask. The Al content of the solution after 2 months was 5.2 μM. The total Al content of the glass, measured after completely dissolving some glass in 48 wt% HF with microwave heating, was 0.80 wt%. What fraction of the Al was extracted from glass by EDTA?

Reference Procedure: Calibrating a 50-mL Buret

This procedure tells how to construct a graph like that in Figure 3-2 (page 66) to convert the measured volume delivered by a buret to the true volume delivered at 20°C.

0. Measure the temperature in the laboratory. Distilled water for this experiment must be at laboratory temperature.

1. Fill the buret with distilled water and force any air bubbles out the tip. See whether the buret drains without leaving drops on its walls. If drops are left, clean the buret with soap and water or soak it with cleaning solution.[5] Adjust the meniscus to be at or slightly below 0.00 mL, and touch the buret tip to a beaker to remove the suspended drop of water. Allow the buret to stand for 5 min while you weigh a 125-mL flask fitted with a rubber stopper. (Hold the flask with a paper towel to prevent fingerprints from changing its mass.) If the level of the liquid in the buret has changed, tighten the stopcock and repeat the procedure. Record the level of the liquid.

2. Drain approximately 10 mL of water at a rate of <20 mL/min into the weighed flask, and cap it tightly to prevent evaporation. Allow 30 s for the film of liquid on the walls to descend before you read the buret. Estimate all readings to the nearest 0.01 mL. Weigh the flask again to determine the mass of water delivered.

3. Drain the buret from 10 to 20 mL, and measure the mass of water delivered. Repeat the procedure for 30, 40, and 50 mL. Then do the entire procedure (10, 20, 30, 40, 50 mL) a second time.

4. Use Table 2-5 to convert the mass of water to the volume delivered. Repeat any set of duplicate buret corrections that do not agree to within 0.04 mL. Prepare a calibration graph as in Figure 3-2, showing the correction factor at each 10-mL interval.

Example **Buret Calibration**

When draining the buret at 24°C, you observe the following values:

Final reading	10.01	10.08 mL
Initial reading	0.03	0.04
Difference	9.98	10.04 mL
Mass	9.984	10.056 g
Actual volume delivered	10.02	10.09 mL
Correction	+0.04	+0.05 mL
Average correction	+0.045 mL	

To calculate the actual volume delivered when 9.984 g of water are delivered at 24°C, use the conversion factor 1.003 8 mL/g in Table 2-5. We find that 9.984 g occupies (9.984 g)(1.003 8 mL/g) = 10.02 mL. The average correction for both sets of data is +0.045 mL.

To obtain the correction for a volume greater than 10 mL, add successive masses of water collected in the flask. Suppose that the following masses were measured:

	Volume interval (mL)	Mass delivered (g)
	0.03–10.01	9.984
	10.01–19.90	9.835
	19.90–30.06	10.071
Sum	30.03 mL	29.890 g

The total volume of water delivered is (29.890 g)(1.003 8 mL/g) = 30.00 mL. Because the indicated volume is 30.03 mL, the buret correction at 30 mL is −0.03 mL.

What does this mean? Suppose that Figure 3-2 applies to your buret. If you begin a titration at 0.04 mL and end at 29.43 mL, you would deliver 29.39 mL if the buret were perfect. Figure 3-2 tells you that the buret delivers 0.03 mL less than the indicated amount; so only 29.36 mL were actually delivered. To use the calibration curve, either begin all titrations near 0.00 mL or correct both the initial and the final readings. Use the calibration curve whenever you use your buret.

Notes and References

1. R. J. Lewis, Sr., *Hazardous Chemicals Desk Reference*, 5th ed. (New York: Wiley, 2002); P. Patnaik, *A Comprehensive Guide to the Hazardous Properties of Chemical Substances*, 2nd ed. (New York: Wiley, 1999); G. Lunn and E. B. Sansone, *Destruction of Hazardous Chemicals in the Laboratory* (New York: Wiley, 1994); and M. A. Armour, *Hazardous Laboratory Chemical Disposal Guide*, 2nd ed. (Boca Raton, FL: CRC Press, 1996).

2. P. T. Anastas and J. C. Warner, *Green Chemistry: Theory and Practice* (New York: Oxford University Press, 1998); M. C. Cann and M. E. Connelly, *Real-World Cases in Green Chemistry* (Washington, DC: American Chemical Society, 2000); M. Lancaster, *Green Chemistry: An Introductory Text* (Cambridge: Royal Society of Chemistry, 2002); C. Baird and M. Cann, *Environmental Chemistry*, 3rd ed. (New York: W. H. Freeman and Company, 2005); J. E. Girard, *Principles of Environmental Chemistry* (Sudbury, MA: Bartlett, 2005); B. Braun, R. Charney, A. Clarens, J. Farrugia, C. Kitchens, C. Lisowski, D. Naistat, and A. O'Neil, *J. Chem. Ed.* **2006**, *83*, 1126.

3. J. M. Bonicamp, *J. Chem. Ed.* **2002**, *79*, 476.

4. Videos illustrating basic laboratory techniques are available from the *Journal of Chemical Education* at http://jchemed.chem.wisc.edu/ and also from www.academysavant.com.

5. Prepare cleaning solution by dissolving 36 g of ammonium peroxydisulfate, $(NH_4)_2S_2O_8$, in a *loosely stoppered* 2.2-L ("one gallon") bottle of 98 wt% sulfuric acid. Add ammonium

peroxydisulfate every few weeks to maintain the oxidizing strength. EOSULF is an alternative cleaning solution for removing proteins and other residues from glassware in a biochemistry lab. EOSULF contains the metal binder EDTA and a sulfonate detergent. It can be safely poured down the drain. [P. L. Manske, T. M. Stimpfel, and E. L. Gershey, *J. Chem. Ed.* **1990**, *67*, A280.]

6. M. M. Singh, C. McGowan, Z. Szafran, and R. M. Pike, *J. Chem. Ed.* **1998**, *75*, 371; *J. Chem. Ed.* **2000**, *77*, 625.

7. R. H. Obenauf and N. Kocherlakota, *Spectroscopy Applications Supplement*, March 2006, p. 12.

8. D. Bohrer, P. Cícero do Nascimento, P. Martins, and R. Binotto, *Anal. Chim. Acta* **2002**, *459*, 267.

Further Reading

H. M. Kanare, *Writing the Laboratory Notebook* (Washington, DC: American Chemical Society, 1985).