Table 1.5 Replicate Determinations of Analyte in a Sample[a]

	% Analyte		
	Analyst 1[b]	Analyst 2[c]	Analyst 3[d]
	10.0	8.1	13.0
	10.2	8.0	10.2
	10.0	8.3	10.3
	10.2	8.2	11.1
	10.1	8.0	13.1
	10.1	8.0	9.3
Average (%)	10.1	8.1	11.2
Absolute error[e]	0.0	2.0	1.1
Standard deviation	0.089	0.13	1.57

[a]Accepted true answer is 10.1 ± 0.2% (obtained independently).
[b]Results are precise and accurate.
[c]Results are precise but inaccurate.
[d]Results are imprecise and inaccurate.
[e]Absolute error = |true value − measured value|.

answer. The latter must be discovered by an independent method, such as having Analyst 2 analyze a sample of known composition. The accepted true value for the determination is 10.1 ± 0.2% according to the table footnote, so the determination by Analyst 2 is not accurate. Analyst 3 is both inaccurate and imprecise. It is very unlikely that an imprecise determination will be accurate. Precision is required for accuracy, but does not guarantee accuracy.

It is important for students to realize that the inability to obtain the correct answer does not necessarily mean that the analyst uses poor laboratory techniques or is a poor chemist. Many causes contribute to poor accuracy and precision, some of which we will discuss in this chapter as well as in later chapters. Careful documentation of analytical procedures, instrument operating conditions, calculations, and final results are crucial in helping the analyst recognize and eliminate errors in analysis.

The quantitative analysis of any particular sample should generate results that are precise and accurate. The results should be reproducible, reliable, and truly representative of the sample. Unfortunately, some degree of error is always involved in analytical determinations, as discussed in Section 1.3.3.

For analytical results to be most useful, it is important to be aware of the reliability of the results. To do this it is necessary to understand the sources of error and to be able to recognize when they can be eliminated and when they cannot. Error is the difference between the true result (or accepted true result) and the measured result. If the error in an analysis is large, serious consequences may result. A patient may undergo expensive and even dangerous medical treatment based on an incorrect laboratory result or an industrial company may implement costly and incorrect modifications to a plant or process because of an analytical error. There are numerous sources of error and several types of errors, some of which are described here.

1.3.3. Types of Errors

There are two principal types of error in analysis: **determinate** or **systematic** error and **indeterminate** or **random** error.

1.3.3.1. Determinate Error

Broadly speaking, **determinate errors** are caused by faults in the analytical procedure or the instruments used in the analysis. The name determinate error implies that the cause of this type of error may be found out and then either avoided or corrected. Determinate errors are *systematic errors*; that is, they are not random. A particular determinate error may cause the analytical results produced by the method to be always too high; another determinate error may render all results too low. Sometimes the error is *constant*; all answers are too high (or too low) by the same amount. If the true results for three samples are 25, 20, and 30 mg/L of analyte, but the measured (or determined) results are 35, 30, and 40 mg/L, respectively, the analysis has a *constant error* of 10 mg/L. Since these results are all too high, the constant error is positive; a constant negative error of 10 mg/L would result in the three measured results being 15, 10, and 20 mg/L of analyte, respectively. Sometimes the determinate error is proportional to the true result, giving rise to **proportional errors**. For example, if the measured results for the same three earlier samples are 27.5, 22.0, and 33.0 mg/L analyte, respectively, the measured results are too high by 10% of the true answer. This error varies in proportion to the true value. Other determinate errors may be variable in both sign and magnitude, such as the change in the volume of a solution as the temperature changes. Although this variation can be positive or negative, it can be identified and accounted for. Determinate errors can be additive or they can be multiplicative. It depends on the error and how it enters into the calculation of the final result.

If you look again at the results in Table 1.5 for Analyst 2, the results produced by this analyst for the repetitive analysis of a single sample agree closely with each other, indicating high precision. However, the results are all too low (and therefore inaccurate), given that Table 1.5 states the true value of the sample to be 10.1 $\pm$ 0.2% analyte. There is a negative determinate error in the results from Analyst 2. This determinate error could be the result of an incorrectly calibrated balance. If the balance is set so that the zero point is actually 0.5 g too high, all masses determined with this balance will be 0.5 g too high. If this balance was used to weigh out the potassium chloride used to make the potassium standard solution used in the clinical laboratory, the standard concentration will be erroneously high, and all of the results obtained using this standard will be erroneously low. The error is reported as the absolute error, the absolute value of the difference between the true and measured values. However, there is not enough information provided to know if this is a constant or a proportional error. It can be seen that close agreement between results (i.e., high precision) does not rule out the presence of a determinate error.

Determinate errors arise from some faulty step in the analytical process. The faulty step is repeated every time the determination is performed. Whether a sample is analyzed 5 times or 50 times, the results may all agree with each other (good precision) but differ widely from the true answer (poor accuracy). An example is given in Table 1.6. Although the replicate results are close to each other, that tells us nothing about their accuracy. We can see from the true value given in Table 1.6 that the experimental results are too high; there is a determinate error in the procedure. An analyst or doctor examining the measured analytical results in Table 1.6 might be deceived into believing that the close agreement among the replicate measurements indicates high accuracy and that the results are close to the true potassium concentration. (Potassium in adult human serum has a normal range of 3.5–5.3 mmol potassium/L serum. Assume that the true value given is for this particular patient.)

In the example in Table 1.6, the true value was 4.0 mmol potassium/L and the average measured result was 5.2 mmol potassium/L in the patient's serum. However,

Table 1.6 Potassium Concentration in a Single Serum Sample

	Measured value (mmol/L)	True value[a] (mmol/L)
	5.2	4.0
	5.1	
	5.3	
	5.1	
	5.1	
Average	5.2	4.0

[a]Normal range for potassium in serum: 3.5–5.3 mmol/L.

the analyst and the doctor do not know the true value of an unknown serum sample. The measured result is in the normal range for adult human serum potassium concentrations, so neither the analyst nor the doctor is likely to be suspicious of the results.

If a faulty analytical procedure is used to analyze five different patients' serum samples and the results shown in Table 1.7 are obtained, it can be seen that in all cases the error is +1.2 mmol/L. This indicates a constant, positive determinate error. As you can see, this faulty procedure would result in one patient being misdiagnosed with a false high serum K level and a patient with a truly low serum K level being misdiagnosed as "normal".

An analyst working at a different hospital with different instrumentation obtains the results shown in Table 1.8. Examination of these analytical results shows they are all ~20% greater than the true answer. The error is *proportional* to the true concentration of the analyte. Such information as to the nature of the error is useful in the diagnosis of the source of the determinate error.

Systematic error is under the control of the analyst. It is the analyst's responsibility to recognize and correct for these *systematic errors* that cause results to be *biased*, that is, offset in the average measured value from the true value. How are determinate errors identified and corrected? Two methods are commonly used to identify the existence of systematic errors. One is to analyze the sample by a completely different analytical procedure that is known to involve no systematic errors. Such methods are often called "standard methods"; they have been evaluated extensively by many laboratories and shown to be accurate and precise. If the results from the two analytical methods agree, it is reasonable to assume that both analytical procedures are free of determinate errors. The second method is to run several analyses of a reference material of known,

Table 1.7 Potassium Concentrations in Patients' Serum

Patient	Measured value[a] (mmol/L)	True value (mmol/L)
A	5.3	4.1
B	4.8	3.6
C	6.3	5.1
D	5.0	3.8
E	4.1	2.9

[a]Constant error of +1.2 mmol/L.

Table 1.8 Potassium Concentration in Serum

Patient	Measured value (mmol/L)	True value[a] (mmol/L)
A	5.8	4.8
B	4.3	3.6
C	7.4	6.2
D	3.5	2.9
E	6.6	5.5

[a]Results indicate a positive proportional error of 20% of the true value.

accepted concentration of analyte. The difference between the known (true) concentration and that measured by analysis should reveal the error. If the results of analysis of a known reference standard are consistently high (or consistently low), then a determinate error is involved in the method. The cause of the error must be identified and either eliminated or controlled if the analytical procedure is to give accurate results. In the earlier example of potassium in serum, standard serum samples with certified concentrations of potassium are available for clinical laboratories. Many clinical and analytical laboratories participate in proficiency testing programs, where "unknown" standard samples are sent to the laboratory on a regular basis. The results of these samples are sent to the government or professional agency running the program. The unknowns are of course known to the agency that sent the test samples; the laboratory receives a report on the accuracy and precision of its performance.

Determinate errors can arise from uncalibrated balances, improperly calibrated volumetric flasks or pipettes, malfunctioning instrumentation, impure chemicals, incorrect analytical procedures or techniques, and analyst error.

Analyst error. The person performing the analysis causes these errors. They may be the result of inexperience, insufficient training, or being "in a hurry". An analyst may use the instrument incorrectly, perhaps by placing the sample in the instrument incorrectly each time or setting the instrument to the wrong conditions for analysis. Consistently misreading a meniscus in a volumetric flask as high (or low) and improper use of pipettes, such as "blowing out" the liquid from a volumetric pipette, are common analyst errors. Some other analyst-related errors are (1) *carelessness*, which is not as common as is generally believed; (2) *transcription errors*, that is, copying the wrong information into a lab notebook or onto a label; and (3) *calculation errors*. Proper training, experience, and attention to detail on the part of the analyst can correct these types of errors.

Reagents and instrumentation. Contaminated or decomposed reagents can cause determinate errors. Impurities in the reagents may interfere with the determination of the analyte, especially at the ppm level or below. Prepared reagents may also be improperly labeled. The suspect reagent may be tested for purity using a known procedure or the analysis should be redone using a different set of reagents and the results compared.

Numerous errors involving instrumentation are possible, including incorrect instrument alignment, incorrect wavelength settings, incorrect reading of values, and incorrect settings of the readout (i.e., zero signal should read zero). Any variation in proper instrument settings can lead to errors. These problems can be eliminated by a systematic procedure to check the instrument settings and operation before use. Such procedures are called standard operating procedures (SOPs) in many labs. There should be a written SOP for each instrument and each analytical method used in the laboratory.

In instrumental analysis, electrical line voltage fluctuations are a particular problem. This is especially true for automated instruments running unattended overnight. Instruments are often calibrated during the day, when electrical power is in high demand. At night, when power demand is lower, line voltage may increase substantially, completely changing the relationship between concentration of analyte and measured signal. Regulated power supplies are highly recommended for analytical instruments. The procedure for unattended analysis should include sufficient calibration checks during the analytical run to identify such problems. Many instruments are now equipped with software that can check the measured value of a standard and automatically recalibrate the instrument if that standard falls outside specified limits.

Analytical method. The most serious errors are those in the method itself. Examples of method errors include (1) incomplete reaction for chemical methods, (2) unexpected interferences from the sample itself or reagents used, (3) having the analyte in the wrong oxidation state for the measurement, (4) loss of analyte during sample preparation by volatilization or precipitation, and (5) an error in calculation based on incorrect assumptions in the procedure (errors can evolve from assignment of an incorrect formula or molecular weight to the sample). Most analytical chemists developing a method check all the compounds likely to be present in the sample to see if they interfere with the determination of the analyte; unlikely interferences may not have been checked. Once a valid method is developed, an SOP for the method should be written so that it is performed the same way every time it is run.

Contamination. Contamination of samples by external sources can be a serious source of error and may be extremely variable. An excellent example of how serious this can be has been documented in the analysis of samples for polychlorinated biphenyls (PCBs). PCBs are synthetic mixtures of organochlorine compounds that were first manufactured in 1929 and have become of concern as significant environmental pollutants. It has been demonstrated that samples archived since 1914, before PCBs were manufactured, picked up measurable amounts of PCBs in a few hours just sitting in a modern laboratory (Erickson). Aluminum levels in the dust in a normal laboratory are so high that dust prohibits the determination of low ppb levels of aluminum in samples. A special dust-free "clean lab" or "clean bench" with a filter to remove small dust particles may be required, similar to the clean rooms needed in the semiconductor industry, for determination of traces of aluminum, silicon, and other common elements such as iron. When **trace** (<ppm level) or **ultratrace** (<ppb level) organic and inorganic analysis is required, the laboratory environment can be a significant source of contamination.

Another major source of contamination in an analysis can be the analyst. It depends on what kind of analytes are being measured, but when trace or ultratrace levels of elements or molecules are being determined, the analyst can be a part of the analytical problem. Many personal care items, such as hand creams, shampoos, powders, and cosmetics, contain significant amounts of chemicals that may be analytes. The problem can be severe for volatile organic compounds in aftershave, perfume, and many other scented products and for silicone polymers, used in many health and beauty products. Powdered gloves may contain a variety of trace elements and should not be used by analysts performing trace element determinations. Hair, skin, and clothing can shed cells or fibers that can contaminate a sample.

Having detected the presence of a determinate error, the next step is to find its source. Practical experience of the analytical method or first-hand observation of the analyst using the procedure is invaluable. Much time can be wasted in an office guessing at the source of the trouble. Unexpected errors can be discovered only in the laboratory. A little data is worth a lot of discussion (Robinson's Law).

1.3.3.2. Indeterminate Error

After all the determinate errors of an analytical procedure have been detected and eliminated, the analytical method is still subject to random or indeterminate error arising from inherent limitations in making physical measurements. Each error may be positive or negative, and the magnitude of each error will vary. Indeterminate errors are not constant or biased. They are random in nature and are the cause of slight variations in results of replicate samples made by the same analyst under the same conditions.

Sources of random error include the limitations of reading balances, scales such as rulers or dials, and electrical "noise" in instruments. For example, a balance that is capable of measuring only to 0.001 g cannot distinguish between two samples with masses of 1.0151 and 1.0149 g. In one case the measured mass is low, in the other case it is high. These random errors cause variation in results, some of which may be too high and some too low, as we see for Analyst 1 in Table 1.5. The average of the replicate determinations is accurate, but each individual determination may vary slightly from the true value. Indeterminate errors arise from sources that cannot be corrected, avoided, or even identified, in some cases. All analytical procedures are subject to indeterminate error. However, because indeterminate error is random, the errors will follow a random distribution. This distribution can be understood using the laws of probability and basic statistics. The extent of indeterminate error can be calculated mathematically.

Let us suppose that an analytical procedure has been developed in which there is no determinate error. If an infinite number of analyses of a single sample were carried out using this procedure, the distribution of numerical results would be shaped like a symmetrical bell (Fig. 1.5). This bell-shaped curve is called the **normal** or **Gaussian distribution**. The frequency of occurrence of any given measured value when only indeterminate error occurs is represented graphically by a plot such as Fig. 1.5.

If only indeterminate errors were involved, the most frequently occurring result would be the true result, that is, the result at the maximum of the curve would be the true answer. In practice it is not possible to make an infinite number of analyses of a single sample. At best, only a few analyses can be carried out, and frequently only one analysis of a particular sample is possible. We can, however, use our knowledge of statistics to determine how reliable these results are. The basis of statistical calculations

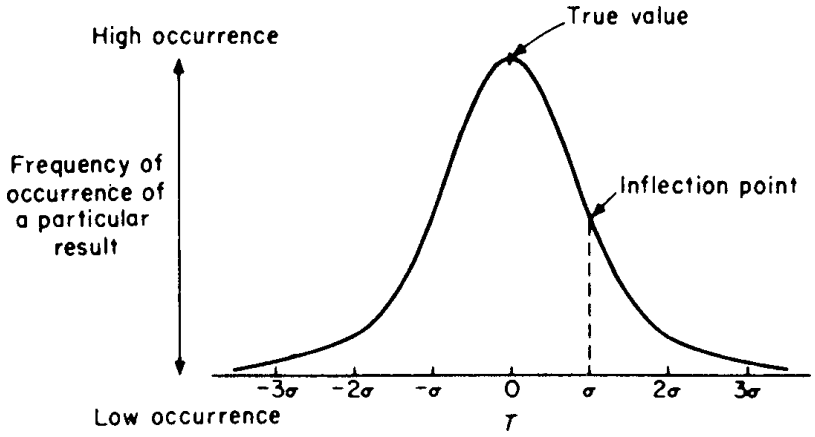

Figure 1.5 A normal or Gaussian distribution of results when only indeterminate error is present. The value that occurs with most frequency is the true value (T) or mean value, while the spread of the distribution is expressed in units of standard deviation from the mean, symbolized by σ. The larger the random error, the broader the distribution will be.

is outlined below. Statisticians differentiate between the values obtained from a finite number of measurements, N, and the values obtained from an infinite number of measurements, so we need to define these statistical terms.

1.3.4. Definitions for Statistics

True value T: the true or accepted value; also symbolized by x_t.
Observed value x_i: a single value measured by experiment.
Sample mean $\bar{x}$: the arithmetic mean of a finite number of observations, that is,

$$\bar{x} = \frac{\sum_{i=1}^{N} x_i}{N} = \frac{(x_1 + x_2 + x_3 + \cdots + x_N)}{N} \tag{1.1}$$

where N is the number of observations and $\sum x_i$ is the sum of all the individual values x_i.

Population mean μ: the limit as N approaches infinity of the sample mean, that is,

$$\mu = \lim_{N \to \infty} \sum_{i=1}^{N} \frac{x_i}{N} \tag{1.2}$$

In the absence of systematic error, the population mean μ equals the true value T of the quantity being measured.

Error E: the difference between the true value T and either a single observed value x_i or the sample mean of the observed values, $\bar{x}$; error may be positive or negative,

$$E = x_i - T \quad \text{or} \quad \bar{x} - x_t \tag{1.3}$$

The total error is the sum of all the systematic and random errors.

Absolute error: the absolute value of E, and can be defined for a single value or for the sample mean,

$$E_{abs} = |x_i - T| \quad \text{or} \quad |\bar{x} - x_t| \tag{1.4}$$

Relative error: the absolute error divided by the true value; it is often expressed as a percent by multiplying by 100,

$$E_{rel} = \frac{E_{abs}}{x_t} \quad \text{or} \quad \%E_{rel} = \frac{E_{abs}}{x_t} \times 100 \tag{1.5}$$

Absolute deviation d_i: the absolute value of the difference between the observed value x_i and the sample mean $\bar{x}$,

$$d_i = |x_i - \bar{x}| \tag{1.6}$$

Relative deviation D: the absolute deviation d_i divided by the mean $\bar{x}$,

$$D = \frac{d_i}{\bar{x}} \tag{1.7}$$

Percent relative deviation: the relative deviation multiplied by 100,

$$D\ (\%) = \frac{d_i \times 100\%}{\bar{x}} = D \times 100 \tag{1.8}$$

Sample standard deviation s: for a finite number of observations N, the sample standard deviation is defined as

$$s = \sqrt{\frac{\sum_{i=1}^{N} d_i^2}{N-1}} = \sqrt{\frac{\sum_{i=1}^{N} (x_i - \bar{x})^2}{N-1}} \tag{1.9}$$

Standard deviation of the mean s_m: the standard deviation associated with the mean of a data set consisting of N measurements,

$$s_m = \frac{s}{\sqrt{N}} \tag{1.10}$$

Population standard deviation σ: for an infinite number of measurements,

$$\sigma = \sqrt{\lim_{N\to\infty} \frac{\sum_{i=1}^{N} (x_i - \mu)^2}{N}} \tag{1.11}$$

Percent relative standard deviation % RSD,

$$\%\ \text{RSD} = \frac{s}{\bar{x}} \times 100 \tag{1.12}$$

Variance σ^2 or s^2: the square of the population standard deviation σ or the sample standard deviation s.

1.3.5. Quantifying Random Error

If the systematic errors have been eliminated, the measured value will still be distributed about the true value owing to random error. For a given set of measurements, how close is the average value of our measurements to the true value? This is where the Gaussian distribution and statistics are used.

The Gaussian distribution curve assumes that an infinite number of measurements of x_i have been made. The maximum of the Gaussian curve occurs at $x = \mu$, the true value of the parameter we are measuring. So, for an infinite number of measurements, the population mean is the true value x_t. We assume that any measurements we make are a subset of the Gaussian distribution. As the number of measurements, N, increases, the difference between $\bar{x}$ and μ tends toward zero. For N greater than 20 to 30 or so, the sample mean rapidly approaches the population mean. For 25 or more replicate measurements, the true value is approximated very well by the experimental mean value. Unfortunately, even 20 measurements of a real sample are not usually possible. Statistics allows us to express the random error associated with the difference between the population mean μ and the mean of a small subset of the population, $\bar{x}$. The random error for the mean of a small subset is equal to $\bar{x} - \mu$.

The area under any portion of the Gaussian distribution, for example, between a value x_1 and a value x_2, corresponds to the fraction of the measurements which will yield a measured value of x between and including these two values. The spread of the Gaussian distribution, that is, the width of the bell-shaped curve, is expressed in terms of the population standard deviation σ. The standard deviation σ coincides with