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COMMISSION ON ANALYTICAL NOMENCLATURE†

GLOSSARY OF BIOANALYTICAL NOMENCLATURE

Part 1: General Terminology, Body Fluids, Enzymology, Immunology

(IUPAC Recommendations 1994)

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Glossary of bioanalytical nomenclature— Part 1: General terminology, body fluids, enzymology, immunology (IUPAC Recommendations 1994)

Synopsis

Many disciplines are involved in the practice of clinical laboratory medicine, with each having its own set of technical terminology. Consequently, the terminology used in the clinical laboratory is often vague, inexact, and, in some cases, even in discord with conventional and officially approved terminology. In order to help rectify this situation, a general set of bioanalytical definitions has been compiled and collated from documents from several national and international organizations with the emphasis on those prepared by the International Federation of Clinical Chemistry (IFCC), the International Union of Pure and Applied Chemistry (IUPAC), and the International Union of Biochemistry (IUB). References are included for each definition included. This document includes sections on General Terminology, Body Fluids, Enzymology, and Immunology. Other topics will be included in subsequent documents. It is hoped that this set of bioanalytical definitions will be useful to the practitioners of clinical chemistry and will foster improved communication and understanding among them.

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As a consequence of being derived from a diversity of disciplines, the terminology used in the clinical laboratory is often vague, inexact, and, in some cases, even in discord with conventional analytical terminology. Consequently, many national and international organizations have prepared and adopted sets of definitions that are pertinent to the field of clinical laboratory science. However, these documents are often prepared for a specific discipline or to meet a specific national need, and it is not unusual for workers in other disciplines to be unaware of them. In addition, these documents are not always easily available, and several are required in order to compile an extensive and comprehensive set of definitions.

In order to help rectify this situation, we have compiled and collated this general set of bioanalytical definitions from documents of several organizations with the emphasis on those prepared by the International Federation of Clinical Chemistry (IFCC), the International Union of Pure and Applied Chemistry (IUPAC), and the International Union of Biochemistry (IUB). References are given for each definition included in this Glossary.

It is hoped that this set of bioanalytical definitions will be useful for the practitioners of clinical chemistry and thus foster improved communication and understanding among them.

1. GENERAL TERMINOLOGY

- 1.1 Accuracy - The closeness of the agreement between the result of a measurement and the (conventional) true value of the measurand (refs. 1,2). See Inaccuracy (1.10). NOTE: The use of the term precision for accuracy must be avoided.
- 1.2 Aliquot - A known fractional portion of a homogeneous material, assumed to be taken with negligible sampling error. The term is usually applied to fluids (ref. 3).
- 1.3 Analyte - The component measured (ref. 3).
- 1.4 Analysis - The determination of the identity of the components of a material or the measurement of their relative or absolute quantity or both (ref. 4).
 - 1.4.1 Qualitative - Identification of the constituents of a material {substance} for certain purposes (ref. 4).

- 1.4.2 Quantitative - The measurement of the relative or absolute quantity and its associated uncertainty (ref. 4).
- 1.5 Bias (of a measuring instrument) - The systematic component of the error of a measuring instrument (ref. 2).
- 1.6 Calibration - The set of operations which established, under specified conditions, the relationship between values indicated by a measuring instrument or measuring system, or values represented by a material measure, and the corresponding known values of a measurand (analyte) (ref. 2).
- NOTES: 1. The result of a calibration permits the estimation of errors of indication of the measuring instrument, measuring system or material measure, or the assignment of values to marks on arbitrary scales.
2. A calibration may also determine other metrological properties.
3. The result of a calibration may be recorded in a document, sometimes called a calibration certificate or a calibration report.
4. The result of a calibration is sometimes expressed as a calibration factor, or as a series of calibration factors in the form of a calibration curve.
- 1.7 Carryover - A process by which materials are carried into a reaction mixture in which they do not belong. This material can be either parts of a sample or reagents including diluent or wash solution. In this case, carryover means transfer of material from one container or from one reaction mixture to another. It can be either unidirectional (backward or forward) or bidirectional in a series of samples or assays (ref. 5).
- 1.8 Error - Difference between a single estimate of a quantity and its true value. This difference deviation (positive or negative) may be expressed either in the units in which the quantity is measured or as a percentage of the true value. If a reliable estimate of the true value is not available, the difference may have to be expressed as the deviation from an assigned value. Note that errors as defined cannot be classified as "random" or "systematic" without considering sets of measurements (see Imprecision, Inaccuracy) (refs. 1,2).
- 1.9 Imprecision - Standard deviation or coefficient of variation of the results in a set of replicate measurements. The mean value and number of replicates must be stated, and the design used must be described in such a way that other workers can repeat it (ref. 1). Using statistical techniques, imprecision can be allocated into separate components (e.g., within run, between day, within series). NOTE: Imprecision also corresponds with the terms "random error" and "experimental standard deviation" as defined by the International Vocabulary of Metrology (ref. 2).
- 1.10 Inaccuracy - Numerical difference between the mean of a set of replicate measurements and true value. This difference (positive or negative) may be expressed in the units in which the quantity is measured or as a percentage of the true value (ref. 1) NOTE: Inaccuracy also corresponds to the term "systematic error" as defined by the International Vocabulary of Metrology (ref. 2).
- 1.11 Interference - The effect of a component on the accuracy of measurement of another component (ref. 1).
- 1.12 Limit of Detection - The smallest single result which, with a stated probability, can be distinguished from a suitable blank (refs. 1,6).
- NOTE: Also corresponds with the term "discrimination threshold" as defined by the International Vocabulary of Metrology (ref. 2).
- 1.13 Measurand - A quantity subjected to measurement (ref. 2).
- NOTE: This term is synonymous with the term analyte (see 1.3).
- 1.14 Method - The set of theoretical and practical operations, in general terms, involved in the performance of measurements according to a given principle (ref. 2). NOTE: See Procedure 1.17.

- 1.14.1 Definitive Method - A method that has been subjected to in-depth investigation and evaluation for sources of inaccuracy. The combined effect of the method imprecision and susceptibility to possible uncorrected systematic error, expressed within the uncertainty statement, if of a magnitude that is compatible with the definitive method's stated end purpose. The midpoint of the overall bounds of error, expressed by the uncertainty statement, may be taken as the true value (refs. 1,7).
- 1.14.2 Reference Method - A method which, after exhaustive investigation, has been shown to have negligible inaccuracy in comparison with its imprecision (ref. 1).
- 1.14.3 Method of Known Bias - A method in which the inaccuracy has been established (for example, by comparison with a reference method) (ref. 1).
- 1.14.4 Method with Unknown Bias - A method whose accuracy is unknown (ref. 1).
- 1.15 Observed Value - The value of a quantity determined as the result of a single observation (ref. 8).
- 1.16 Precision - The closeness of agreement between independent test results obtained under prescribed conditions (refs. 1,8). (See Imprecision, 1.9.)
- 1.17 Procedure - The set of theoretical and practical operations, in detailed terms, involved in the performance of measurements according to a given method (ref. 2). NOTE: See Method 1.14.
- 1.18 Range, Analytical - The range of concentration or other quantity values in the specimen over which the method is applicable without modification (ref. 1).
- 1.19 Reference Material - A material or substance, one or more properties of which are sufficiently well established to be used for the calibration of an apparatus, the assessment of a measurement method, or for assigning values to materials (refs. 2,8,9).
- 1.19.1 Certified Reference Material (CRM) - A reference material, one or more of whose property values are certified by technically valid procedure accompanied by or traceable to a certificate or other documentation which is issued by a certifying body (refs. 2,8,9).
- 1.19.2 Calibration Material - A reference material used in an analytical process to assign values to specimens by relating the readings or analytical responses obtained to the concentration or other quantities present (refs. 1,10).
- 1.19.2.1 Primary Calibration Material - The term reserved for thoroughly characterized, stable, homogeneous materials of which one or more physical or chemical properties have been experimentally determined within stated measurement uncertainties (ref. 10).
- 1.19.2.2 Secondary Calibration Material - A material, that is traceable to a primary calibration material, which includes pure substances and materials in a matrix that reproduces or simulates the expected matrix (ref. 10).
- 1.19.3 Control Materials - Essentially identical specimens of stable materials, usually similar in composition and physical properties to natural specimens, carried through the same analytical method for surveillance control (monitoring) to estimate performance characteristics (precision, accuracy, etc.) of the analytical process (ref. 11). Such materials are analyzed solely for quality control purposes, not for calibration (ref. 1).
- 1.19.4 Value - A value attached to a calibration material or a control material is a number, together with the name or symbol of its unit. It is applied with respect to a specific batch or lot of a calibration material. The value should be accompanied by a statement expressing the confidence limits or uncertainty associated with that value (refs. 10,11).

- 1.19.5 Principal Assigned Value - A value that has been assigned to a calibration material or a control material by the use of a definitive method, a reference method, or an otherwise officially designated or thoroughly-defined analytical method. A calibration material or control material assigned a principal assigned value is intended to represent a true value insofar as the state of the preparative and analytical art permits (refs. 10,11).
- 1.19.6 Specific System Value - A value that has been assigned to a calibration material or control material intended for or recommended for use only with the analytical systems that are specified (refs. 10,11).
- 1.20 Repeatability - The closeness of the agreement between the results of successive measurements of the same measurand carried out subject to all of the following conditions:
- the same method of measurement
 - the same observer
 - the same measuring instrument
 - the same location
 - the same conditions of use
 - repetition over a short period of time (refs. 2,8).
- NOTE: Repeatability may be expressed quantitatively in terms of the dispersion of the results.
- 1.21 Reproducibility - The closeness of agreement between the results of measurements obtained with the same method of identical material in different laboratories with different operators using different equipment. (See Imprecision, 1.9).
- method of measurement
 - observer
 - measuring instrument
 - location
 - conditions of use
 - time (refs. 2,8).
- NOTES: 1. A valid statement of reproducibility requires specification of the conditions changed.
2. Reproducibility may be expressed quantitatively in terms of the dispersion of the results.
- 1.22 Result - Final value obtained for a measured quantity after performing a measuring procedure including all subprocedures and laboratory evaluations (refs. 1,2,8).
- 1.23 Run - A set of measurements performed by an analytical instrument without interruption (ref. 12).
- 1.24 Sample - A portion of material selected from a larger quantity of material (ref. 3).
- 1.25 Specificity - The ability of an analytical method to determine solely the component(s) it purports to measure. It is assessed on the evidence available on the components which contribute to the result, and on the extent to which they do (ref. 1).
- 1.26 Specimen - A specifically selected portion of a material taken from a dynamic system and assumed to be representative of the parent material at the time it is taken (ref. 3).
- 1.27 Value, True - Term considered to have a self-evident meaning requiring no definition. In practice, the true value is the principal assigned value defined by a definitive method and somewhat less closely by the principal assigned value defined by a reference method (ref. 1).
- 1.28 Value, Conventional True - A value of a quantity which, for a given purpose, may be substituted for the true value (ref. 2). NOTE: A conventional true value is, in general, regarded as sufficiently close to the true value for the difference to be insignificant for the given purpose (ref. 2).

BODY FLUIDS

The clinical laboratory receives samples of many different types of body fluids for analysis. Definitions are included here for the more common types of samples that are routinely encountered in the clinical laboratory.

- 2.1 Blood - The fluid that circulates through the heart, arteries, capillaries, and veins. Blood is composed of plasma, the fluid portion, and cells, the particles suspended in the plasma ([ref. 4](#)).
- 2.1.1 Blood Cell - Any cellular element of the blood including erythrocytes, leukocytes, and platelets ([ref. 4](#)).
- 2.1.2 Blood Cell Count - The number of red blood cells and white blood cells per unit volume in a specimen of venous blood. In some instances, blood cell count may also include measurement of the hematocrit, hemoglobin, and various computed erythrocyte indices (mean corpuscular volume; mean corpuscular hemoglobin; mean corpuscular hemoglobin concentration) ([ref. 4](#)).
- 2.1.3 Blood Group - Red cell phenotypes classified by their antigenic structural characteristics, which are under the control of various allelic genes. The cell membrane properties that provide the specific antigenicity of the blood groups are called agglutinogens as they agglutinate or clump in the presence of their specific antibody ([ref. 4](#)).
- 2.1.4 Cord Blood - The blood contained in the vessels of the umbilical cord at the time of birth ([ref. 4](#)).
- 2.1.5 Occult Blood - Blood present in such small amounts that its presence can be ascertained only by chemical analysis or by spectroscopic or microscopic examination; particularly the blood found in stools ([ref. 4](#)).
- 2.1.6 Peripheral Blood - Blood obtained from parts of the body that are located at some distance from the heart. Examples are blood drawn from the earlobe, fingertip, or heel pad ([ref. 4](#)).
- 2.1.7 Plasma - A clear, yellowish fluid that accounts for about 55 percent of the total volume of blood. Plasma is obtained by centrifuging a whole blood sample that has had an anticoagulant added to it. Plasma from which fibrinogen and related coagulation proteins have been removed is called serum ([ref. 4](#)).
- 2.1.8 Serum - The clear, yellowish fluid that separates from blood when it is allowed to clot. It closely resembles plasma except for the absence of some coagulation factors ([ref. 4](#)).
- 2.1.9 Protein-Free Filtrate - A sample of blood, serum, or plasma from which all proteins have been removed by chemical or physical denaturation, dialysis, ultrafiltration, or solvent extraction ([ref. 4](#)).
- 2.2 Urine - A fluid, containing water and metabolic products, that is excreted by the kidneys, stored in the bladder, and normally discharged by way of the urethra ([ref. 4](#)).
- 2.3 Miscellaneous Body Fluids
 - 2.3.1 Amniotic Fluid - The fluid that surrounds the fetus in the amniotic sac. A specimen is obtained by a technique called amniocentesis in which a long needle is inserted into the amniotic sac through the abdominal wall through which fluid is withdrawn ([ref. 4](#)).
 - 2.3.2 Cerebrospinal Fluid (CSF) - A clear, colorless fluid that fills spaces within and around the central nervous system. It is formed from plasma by a biological ultrafiltration process. Specimens are obtained by a lumbar puncture (i.e., a spinal tap) ([ref. 4](#)).
 - 2.3.3 Lymph - A yellowish, slightly basic fluid derived from tissue fluid. Lymph is collected from peripheral tissues throughout the body and is carried in lymph vessels to the circulatory system via the thoracic duct and the right lymphatic duct ([ref. 4](#)).

- 2.3.4 Saliva - The clear, viscous secretion from the parotid, submaxillary, sublingual, and smaller mucous glands in the cavity of the mouth ([ref. 4](#)).

3. ENZYMOLOGY

3.1 General Terminology

- 3.1.1 Activation Energy - An operationally defined quantity expressing the dependence of a rate coefficient on temperature ([ref. 13](#)). In enzymology it is usually considered to be the energy required for a molecule to form an activated complex which is in the transition of making or breaking a chemical bond. In an enzyme-catalyzed reaction, this corresponds to the formation of the activated enzyme-substrate complex ([refs. 4,21](#)).
- 3.1.2 Activator - An effector molecule that increases the catalytic activity of an enzyme when it binds to a specific site ([ref. 4](#)).
- 3.1.3 Active Center - The term is often applied to the sites that are the effective sites for a particular heterogeneous catalytic reaction ([ref. 13](#)). In enzymology it is considered to be that part of enzyme or other protein at which the initial binding of substrate and enzyme occurs to form the intermediate enzyme-substrate complex and at which location further chemical change characteristic of the catalyzed reaction takes place ([ref. 14](#)).
- 3.1.4 Allostery - A phenomenon whereby the conformation of an enzyme or other protein is altered by combination, at a site other than the substrate-binding site, with a small molecule, referred to as an effector, which results in either increased or decreased activity by the enzyme ([ref. 4](#)).
- 3.1.5 Apoenzyme - The protein part of an enzyme without the cofactor necessary for catalysis. The cofactor can be a metal ion, an organic molecule (coenzyme), or a combination of both ([ref. 4](#)).
- 3.1.6 Catalyst - A catalyst is a substance that increases the rate of a reaction without modifying the overall standard Gibbs energy change in the reaction; the process is called catalysis, and a reaction in which a catalyst is involved is known as a catalyzed reaction ([refs. 13,19](#)). NOTE: An enzyme is a biocatalyst.
- 3.1.7 Catalytic Activity - The property of a catalyst which is measured by the catalyzed rate of conversion of a specified chemical reaction, produced in a specified assay system ([refs. 15,16,17,21](#)).
- 3.1.8 Catalytic Activity Concentration - The property of an enzyme obtained by dividing the catalytic activity by the volume of the original system from which the sample comes ([refs. 1,15](#)).
NOTE: In actual practice, the name may be shortened to "catalytic concentration."
- 3.1.9 Coenzyme - The dissociable, low-relative-molecular-mass active group of an enzyme which transfers chemical groups, hydrogen, or electrons. A coenzyme binds with its associated protein (apoenzyme) to form the active enzyme (holoenzyme) ([refs. 4,14](#)).
- 3.1.10 Denaturation - The partial or total alteration of the structure of a protein without change in covalent structure by the action of certain physical procedures (heating, agitation) or chemical agents. Denaturation is the result of the disruption of tertiary bonding, which causes the opening of the folded structure of a protein and the loss of characteristic physiologic, enzymatic, or physicochemical properties; it can be either reversible or irreversible ([ref. 4](#)).
- 3.1.11 Enzyme - An enzyme is a protein that acts as a catalyst ([ref. 18](#)).
- 3.1.12 Holoenzyme - An active enzyme consisting of the apoenzyme and coenzyme.

- 3.1.13 Immobilized Enzymes - Soluble enzymes bound to an insoluble organic or inorganic matrix, or encapsulated within a membrane in order to increase their stability and make possible their repeated or continued use (ref. 14).
- 3.1.14 Induction - In enzymology, induction is a biological process which results in an increased biosynthesis of an enzyme thereby increasing its apparent activity. It results from the presence of an inducer (ref. 4).
- 3.1.15 Inhibition - An inhibitor is a substance that diminishes the rate of a chemical reaction; the process is called inhibition (refs. 13,18,19).
NOTE: For a further subclassification of types of inhibition, the reader is directed to ref. 18.
- 3.1.16 Isoenzyme - One of a group of related enzymes catalyzing the same reaction but having different molecular structures and characterized by varying physical, biochemical, and immunological properties (ref. 4).
- 3.1.17 Lineweaver-Burk Plot - A Lineweaver-Burk plot is a plot of the reciprocal of velocity of an enzyme-catalyzed reaction (ordinate) versus the reciprocal of substrate concentration (abscissa). The plot is used to graphically define the maximum velocity of an enzyme-catalyzed reaction and the Michaelis constant for the enzyme (ref. 4).
- 3.1.18 Michaelis-Menten Kinetics - Sometimes the relationship between the rate of an enzyme-catalyzed reaction v and the substrate concentration $[A]$ takes the form

$$v = \frac{V[A]}{K_{mA} + [A]}$$

where V and K_{mA} are constants at a given temperature and a given enzyme concentration. The reaction is then said to display Michaelis-Menten kinetics. (The term hyperbolic kinetics is also sometimes used because a plot of v against $[A]$ has the form of a rectangular hyperbola through the origin with asymptotes $v = V$ and $[A] = -K_{mA}$). This term, and others that imply the use of particular kinds of plot, should be used with care to avoid ambiguity, as they can be misleading if used out of context.

NOTE: The quantity $k_0[E]_0$ is given the symbol V and the name *limiting rate*. It is particularly useful when k_0 cannot be calculated because the total catalytic-center concentration is unknown, as in studies of enzymes of unknown purity, sub-unit structure and molecular mass. The symbol V_{max} and the names *maximum rate* and *maximum velocity* are also in widespread use although under normal circumstances there is no finite substrate concentration at which $v = V$ and hence no maximum in the mathematical sense. The form V_{max} is convenient in speech as it avoids the need for a cumbersome distinction between "capital V " and "lower case v ". When a true maximum does occur (as in substrate inhibition: the symbol v_{max} (not V_{max}) and the name maximum rate may be used for the true maximum value of v but care should be taken to avoid confusion with the limiting rate.)

The second constant K_{mA} is known as the Michaelis constant for A; the alternative name Michaelis concentration may also be used and has the advantage of emphasizing that the quantity concerned has the dimensions of a concentration and is not, in general, an equilibrium constant. When only one substrate is being considered, the qualifier A may be omitted, so that the symbol becomes K_m .

When the qualifier is included, its location is a matter of typographical convenience; no particular significance attaches to such variants as K_m or K_{mA} . The Michaelis constant (or Michaelis concentration) is the substrate concentration at which $v = 0.5 V$, and its usual unit is mol dm⁻³ which may be written as mol L⁻¹ (ref. 21). The term Michaelis constant and the symbol K_m should not be used when Michaelis-Menten kinetics are not obeyed (ref. 18).

- 3.1.19 Product - The substance produced by the enzyme-catalyzed conversion of a substrate ([ref. 4](#)).
- 3.1.20 Substrate - A substrate is a reactant (other than the catalyst itself) in a catalyzed reaction ([refs. 18,19](#)).

3.2 Types of Kinetic Conditions

- 3.2.1 Pseudo-Zero-Order Reaction - A zero-order reaction is one in which the rate of reaction is independent of the concentration of reactant. A reaction may be described as pseudo-zero order if it is independent of the reaction of the particular substrate being varied (e.g., the concentration of the enzyme is held constant). This may apply when the enzyme is saturated with substrate over the range of substrate concentration studied ([refs. 15,18,19](#)).
- 3.2.2 Pseudo-First-Order Reaction - A first-order reaction is one in which the rate of reaction is proportional to the concentration of reactant. A second-order reaction, whose rate is proportional to the product of the concentrations of two reactants, may be described as pseudo-first order when the concentration of one is held constant, so that its rate is directly proportional to the concentration of the other ([refs. 15,18,19](#)).

3.3 Units Used to Express Enzyme Activity

- 3.3.1 International Unit - The unit enzyme activity proposed by the International Union of Biochemistry in 1964. Specifically, it is the amount of enzyme that catalyzes the conversion of one micromole of substrate per minute under the specified conditions of the assay method ([ref. 4](#)).

NOTE: This unit is no longer recommended because the term does not indicate what physical quantity it refers to, and because the minute is not the SI unit of time.

- 3.3.2 Katal - The amount of enzyme activity that converts one mole of substrate per second under specified reaction conditions ([refs. 13,24](#)).

NOTE: The katal is now the recommended unit to express enzyme activity ([ref. 1](#)). It is also called the mole per second ([ref. 16](#)).

3.4 Measurement Modes

- 3.4.1 One Point - A reaction mode in which only one measurement is made during the progress of the reaction ([ref. 15](#)). The measurement can be made either after stopping the reaction or when the reaction is in progress ([ref. 20](#)).

- 3.4.2 Two Point - A reaction mode in which individual measurements are taken at time t_1 and t_2 . These times may either be fixed or variable.

3.4.2.1 Fixed Time - A two-point reaction mode in which measurements are taken at specified (i.e., "fixed") times. This mode is preferred for assays in which the reaction rate is first order in regard to the initial substrate concentration ([refs. 15,20](#)).

3.4.2.2 Variable Time - A two-point reaction mode in which the first measurement is taken at a specified time (t_1) and the second at a time after which a fixed signal change has occurred. This mode is preferred for the determination of the catalytic activity concentration of enzymes or other catalysts ([refs. 15,20](#)).

- 3.4.3 Multipoint - A reaction mode in which at least three data points are obtained during the time course of the reaction ([ref. 20](#)).

3.4.3.1 Sequential Two Point - A reaction mode in which a series of data points are accumulated by measuring the signal changes occurring in an assigned sequence of time intervals (consecutive or overlapping). Various algorithms can

then be used to convert the series of intervals of signal change into units of enzyme activity (ref. 20).

3.4.3.2 Multipoint with Average Rate - A reaction mode in which enzyme activity is determined by either averaging the rate for each interval measured or averaging selected intervals after some have been rejected (ref. 20).

3.4.3.3 Multipoint with Computer Analysis - A reaction mode in which multiple points are obtained and the rate obtained by applying a mathematical model to data (ref. 20).

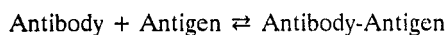
3.4.4 Continuous - A reaction mode in which the reaction is monitored continuously and the data presented in either an analog or digital mode (refs. 15,20).

4. IMMUNOLOGY

4.1 Adjuvant - A material introduced with an antigen to augment its immunogenicity (ref. 22).

4.2 Adsorb - To collect on a surface usually by a monomolecular layer of reactant. Materials such as plastic, glass, or particles (latex, bentonite, cellulose, etc.) that are used for the removal of antibodies or antigens by immobilizing the appropriate reactant to their surface (ref. 22).

4.3 Affinity - Defines and quantitates the force of association between a single antibody and a single epitope of antigen; the association constant, K , for the immunochemical reaction:



which can be expressed mathematically (ref. 22) as:

$$K = \frac{[\text{Antibody-Antigen}]}{[\text{Antibody}][\text{Antigen}]}$$

4.4 Average Affinity Constant(K) - The average binding constant (K) of a population of antibody molecules (ref. 22).

4.5 Agglutination - The immunochemical-specific aggregation of particulate matter such as bacteria, erythrocytes, or other cells, or synthetic particles such as plastic beads coated with antigens or antibodies. Such aggregation usually is dependent primarily on surface reactions mediated either by antigens or by antibodies physically or chemically attached to the particulate surfaces; agglutination of clumping of the particles follows as a secondary immune reaction (ref. 22).

4.5.1 Agglutination Inhibition - A type of agglutination in which particulate and soluble antigen compete for soluble antibody. Soluble antigen in the test medium reacts first with the soluble antibody, inhibiting agglutination of indicator particles. With viral hemagglutination inhibition assays, host antibodies resulting from a specific infection are the most common forms of agglutination inhibition assays. In this case, viral-specific antibodies block the sites on the virus that agglutinate erythrocytes (ref. 22).

4.5.2 Hemagglutination - Agglutination reactions in which the particles used are erythrocytes. Hemagglutination may be either direct, in which erythrocyte antigens are reactants, or indirect (passive) for coated antigen or, in the case of reverse (passive) assays, coated antibody. One of the most common uses of hemagglutination is to quantitate the number of hemagglutinating viruses (cf. influenza) or their soluble hemagglutinating surface subunits (ref. 22).

4.5.3 Indirect (Passive) Agglutination - The agglutination technique in which antigen first is coated artificially onto the particulate surfaces, either by physical absorption or chemical or immunochemical linkage. These antigen-laden particles then can be used to detect the presence of the corresponding specific agglutinins in test material. Agglutination results by cross-linking of the antigen-bearing particles onto an extensive antigen-antibody lattice (i.e., in detectable agglutination of the particles) (ref. 22).

- 4.5.4 Mixed Agglutination Tests - The mixed agglutination phenomenon results from the reaction of antibody with similar antigenic determinants on two different types of particles (or cell types), with aggregation of these disparate particles. One particle is the indicator system for antibodies directed against and bound to the other particle or its surface antigen ([ref. 22](#)).
- 4.5.5 Reverse Passive Agglutination - In this modification of the indirect agglutination assay, particles are coated with antibody. These antibody-laden particles then become probes for detecting specific antigens in test material. Presence of the relevant antigen will result in agglutination of particles ([ref. 22](#)).
- 4.6 Antibody - The functional component of antiserum, often referred to collectively as a population of molecules, each member of which is capable of reacting with a specific antigenic determinant. An antibody molecule is, by definition, monospecific but might also be "idiospecific," "heterospecific," "polyspecific," or of "unwanted specificity." It cannot be "nonspecific" except in the sense of nonimmunochemical binding. These proteins are immunoglobulins and bind by means of specific binding sites to a specific antigenic determinant ([ref. 22](#)).
- 4.7 Antigen - Classically, a substance that will elicit the formation of antibodies in a suitable host. A more recent connotation defines an antigen as a substance that will combine with antibody through its antibody-binding sites ([ref. 22](#)).
- 4.8 Antigenic Determinant - That part of the structure of an antigen molecule that is responsible for specific interaction with antibody molecules evoked by the same or a similar antigen ([ref. 22](#)).
- 4.9 Antiserum - A serum containing antibodies ([ref. 22](#)).
- 4.10 Avidity - Operationally defines the combined intensity of reactivities of an antiserum or antibody population ([ref. 22](#)). In effect, it represents the net affinity of all binding sites of all antibodies in the antiserum, under specified physicochemical reaction conditions. The avidity is a function of the affinities of the antibody-combining sites on all antibodies present in an antiserum and all the antigenic determinants of available macromolecules. Sometimes avidity can be expressed as an effective affinity constant (see 4.3).
- 4.11 Binding Capacity - The capacity of a receptor to bind a ligand, expressed in operational units, unlike the quantitative mass units of the affinity constant ([ref. 22](#)).
- 4.12 Bound/Free (*B/F*) Ratio - The ratio of bound to free-labeled analyte in an immunoassay ([ref. 22](#)).
- 4.13 Carrier Protein - A protein to which a specific ligand or hapten (see 4.19) is conjugated. Also refers to unlabeled protein introduced into an assay at relatively high concentrations which distributes in a fractionation process in the same manner as a labeled protein analyte, present in very low concentrations. Also, a protein added to prevent nonspecific interaction of reagents with surfaces, sample components, and each other ([ref. 22](#)).
- 4.14 Complement - An array of serum proteins (some of which are enzymes that become sequentially activated after the first member of the series is activated by either antigen-antibody complexes or microbial products ([ref. 22](#)).
- 4.15 Conjugate - A material produced by attaching two or more substances together. Conjugates of antibody with fluorochromes, radioactive isotopes, or enzymes are often used in immunoassays ([ref. 22](#)).
- 4.16 Cross Reactivity - The reaction of an antibody with an antigen other than that which elicited the formation due to the presence of related determinants ([ref. 22](#)).
- 4.17 Enzyme Conjugate - Designates a material that has an enzyme bound covalently ([ref. 22](#)).
- 4.18 Epitope - The minimum molecular structure ("antigenic determinant") that will react with an antibody and may be only a portion of antigen ([ref. 22](#)).

- 4.19 Hapten - A specific substance that interacts with specific antibody-combining sites of an antibody molecule, but is not immunogenic by itself (ref. 22).
- 4.20 Heterogeneous Immunoassay - An immunoassay that requires the physical separation of free-labeled antigen (or antibody) from labeled antigen (or antibody) bound in an immune complex, prior to measurement of the quantity of label (ref. 22).
- 4.21 Homogeneous Immunoassay - An immunoassay in which no physical separation is performed. The specific activity of the label or the signal is modulated according to the analyte content of the sample (ref. 22).
- 4.22 Hybridoma - A cell derived from the fusion between a B cell and a plasmacytoma (ref. 22).
- 4.23 Immunoassay - A ligand-binding assay that uses a specific antigen or antibody capable of binding to the analyte (ref. 22).
- 4.24 Immunogen - A substance that elicits a cellular immune response and/or antibody production (see 4.6) (ref. 22).
- 4.25 Immunogenicity - The ability of an immunogen to elicit an immune response (ref. 22).
- 4.26 Immunoglobulin - A glycoprotein found in serum or other body fluids possessing antibody activity (ref.22).
- 4.26.1 Immunoglobulin Class - A classification of immunoglobulin based on antigenic and structural differences of the Heavy Peptide Chain. There are five classes: IgG, IgA, IgM, IgD, and IgE (ref. 22).
- 4.26.2 Immunoglobulin Subclass - A subdivision of the classes based on structural and antigenic differences in the Heavy Peptide Chain; Isotype. Four human IgG subclasses and two IgA subclasses have currently been recognized; IgM subclasses have been postulated; IgD and IgE subclasses are unknown (ref. 22).
- 4.27 Immunoprecipitin Analysis - Any analysis that relies on a system that analyzes a precipitate formed between antibody and antigen (ref. 22).
- 4.28 Label - An easily detected substance that is attached to a reagent in an immunoassay. The assay signal is either a measurable property of the label or is produced by the label. In enzyme immunoassay (EIA) (see 4.33.1) the label is the enzyme; in fluorescence immunoassay (FIA) (see 4.33.3) the label is a fluorescent material; in radioimmunoassay (RIA) (see 4.36.2) the label is the radionuclide (ref. 22).
- 4.29 Ligand - A substance or part of a substance that binds to a specific receptor (ref. 22).
- 4.30 Matrix - The milieu of the sample (e.g., serum) containing the analyte. The matrix can influence the behavior of an immunoassay due to specific (direct) and nonspecific (indirect) interferences (ref. 22).
- 4.31 Monoclonal - Arising from a single clone of cells, in the case of immunoglobulin, refers to its origin; usually the monoclonal antibody is of a single immunoglobulin class containing only one light chain type of either the K or L variety. Also refers to all antibody molecules having identical physical-chemical characteristics and antibody specificity. Monoclonal antibodies have very restricted structural diversity and are homogeneous compared with polyclonal antibodies (ref. 22).
- 4.32 Monospecificity - Monospecificity is functionally defined as the immunoreactivity of an antiserum with its designated antigen (e.g., antihuman IgG, antihuman IgG Fc piece, human IgG3 Fc piece, etc.). In practice, true monospecificity to naturally occurring antigens does not occur in antisera produced by the immunization of the intact animal. An attempt is made to reduce the level of unwanted specificities below that which will interfere with the intended use of a particular immunochemical test (ref. 22).

- 4.33 Nonradioisotopic Immunoassay - A type of immunoassay in which the antigen-antibody reaction is measured through the light-scattering properties of immune complexes or through the use of marker molecules attached to constituents of the immune reaction ([ref. 22](#)).
- 4.33.1 Enzyme Immunoassay (EIA) - A generic term for an immunoassay in which the analyte content of the sample is estimated by measuring the catalytic activity of a specific enzyme conjugate on a substrate ([ref. 22](#)).
- 4.33.2 Enzyme-Linked Immunosorbent Assay (ELISA) - A heterogeneous enzyme immunoassay method where an antigen or antibody is firmly attached to a solid support ([ref. 22](#)).
- 4.33.3 Fluorescence Immunoassay (FIA) - A generic term for an immunoassay in which the analyte is measured by fluorescence. This type of assay is carried out by conjugating fluorescent compounds to the antigen or antibody and then measuring the fluorescence in the antigen-antibody reaction ([refs. 4,22](#)).
- 4.33.4 Light-Scattering Immunoassay - A type of immunoassay which involves the detection of the antigen-antibody complex formation in an immune reaction by changes in turbidity (turbidimetry) or light scattering (nephelometry) in a fluid medium ([ref. 4](#)).
- 4.34 Polyclonal - Arising from different clones ([ref. 22](#)). A typical antiserum obtained from a conventional immunization is polyclonal.
- 4.35 Potency - The characteristic of an antibody representing the concentration (titre) of antibody and the avidity for a given substrate (antigen) in the defined method ([ref. 22](#)).
- 4.36 Radioligand Assay - A technique in which unlabeled and radioactive labeled molecules of the same species compete for a limited number of binding sites on a specific binding protein. The binding protein may be an antibody, transport protein, hormone receptor, or any other cell-associated receptor or tissue component. The unlabeled ligand is the analyte. In the procedure, after a suitable reaction period, the bound ligand (both labeled and unlabeled) is separated from the free ligand, and the radioactivity of either fraction is measured. Calibration reference materials are included in the assay, and the concentration of unlabelled ligand can be estimated from the calibration curve or computed after application of a suitable curve-fitting routine ([ref. 4](#)).
- 4.36.1 Competitive Protein Binding Assay - A type of radio-ligand assay in which the binding protein is a transport protein or enzyme ([ref. 4](#)).
- 4.36.2 Radioimmunoassay (RIA) - A type of radioligand assay in which the binding protein is an antibody ([ref. 4](#)).
- 4.36.3 Radioreceptor Assay - A type of radioligand assay in which the binding protein is a hormone receptor ([ref. 4](#)).
- 4.37 Receptor - A specific molecule on the surface of a neuron or target cell that specifically binds to a specific molecule such as a neurotransmitter, hormone, antigen complement component, lymphokine, etc. ([ref. 4](#)).
- 4.38 Sandwich Immunoassay - An immunoassay using the chemical or immunochemical binding of the analyte to a solid phase and the immunochemical binding of a second (labeled) reagent to the analyte ([ref. 22](#)).
- 4.39 Specificity - An antiserum quality defining its reactivity with defined antigens. In a chemical context, the extent to which the assay responds only to (all subsets of) a specified analyte and not to other substances present in the sample ([ref. 22](#)).
- 4.40 Titre - The reciprocal of the dilution factor required to produce a defined outcome to a defined system. In a defined system it is usually proportional to the analyte concentration ([ref. 22](#)).

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APPENDIX

ALPHABETICAL LISTING OF TERMS

(Numbers in parenthesis refers to sections)

Accuracy (1.1)	Bound/Free (B/F) Ratio (4.12)
Activation Energy (3.1.1)	Calibration (1.6)
Activator (3.1.2)	Calibration Material (1.19.2)
Active Centre (3.1.3)	Carrier Protein (4.13)
Adjuvant (4.1)	Carryover (1.7)
Adsorb (4.2)	Catalyst (3.1.6)
Affinity (4.3)	Catalytic Activity (3.1.7)
Agglutination (4.5)	Catalytic Activity Concentration (3.1.8)
Agglutination Inhibition (4.5.1)	Cerebrospinal Fluid (2.3.2)
Aliquot (1.2)	Certified Reference Material (1.19.1)
Allostery (3.1.4)	Coenzyme (3.1.9)
Amniotic Fluid (2.3.1)	Competitive Protein Binding Assay (4.36.1)
Analysis (1.4)	Complement (4.14)
Analyte (1.3)	Conjugate (4.15)
Antibody (4.6)	Continuous (3.4.4)
Antigen (4.7)	Control Materials (1.19.3)
Antigenic Determinant (4.8)	Cord Blood (2.1.4)
Antiserum (4.9)	Cross Reactivity (4.16)
Apoenzyme (3.1.5)	Definitive Method (1.14.1)
Average Affinity Constant (4.4)	Denaturation (3.1.10)
Avidity (4.10)	Enzyme (3.1.11)
Bias (1.5)	Enzyme Conjugate (4.1)
Binding Capacity (4.1)	Enzyme General Terminology (3.1)
Blood (2.1)	Enzyme Immunoassay (4.33.1)
Blood Cell (2.1.1)	Enzyme-Linked Immunosorbent Assay (4.33.2)
Blood Cell Count (2.1.2)	Enzymology (3.0)
Blood Group (2.1.3)	Epitope (4.18)
Body Fluids (2.0)	Error (1.8)

ALPHABETICAL LISTING OF TERMS
(continued)

Fixed Time (3.4.2.1)	Light-Scattering Immunoassay (4.33.4)
Fluorescence Immunoassay (4.33.3)	Limit of Detection (1.12)
General Terminology (1)	Lineweaver-Burk Plot (3.1.17)
Hapten (4.19)	Lymph (2.3.3)
Hemagglutination (4.5.2)	Matrix (4.30)
Heterogeneous Immunoassay (4.20)	Measurand (1.13)
Holoenzyme (3.1.12)	Measurement Modes (3.4)
Homogeneous Immunoassay (4.21)	Method (1.14)
Hybridoma (4.22)	Method of Known Bias (1.14.3)
Immobilized Enzymes (3.1.13)	Method of Unknown Bias (1.14.4)
Immunoassay (4.23)	Michaelis-Menten Kinetics (3.1.18)
Immunogen (4.24)	Miscellaneous Body Fluids (2.3)
Immunogenicity (4.25)	Mixed Agglutination Tests (4.5.4)
Immunoglobulin (4.26)	Monoclonal (4.31)
Immunoglobulin Class (4.26.1)	Monospecificity (4.32)
Immunoglobulin Subclass (4.26.2)	Multipoint (3.4.3)
Immunology (4)	Multipoint with Average Rate (3.4.3.2)
Immunoprecipitin Analysis (4.27)	Multipoint with Computer Analysis (3.4.3.3)
Imprecision (1.9)	Nonradioisotopic Immunoassay (4.33)
Inaccuracy (1.10)	Observed Value (1.15)
Indirect (Passive) Agglutination (4.5.3)	Occult Blood (2.1.5)
Induction (3.1.14)	One Point (3.4.1)
Inhibition (3.1.15)	Peripheral Blood (2.1.6)
Interference (1.11)	Plasma (2.1.7)
International Unit (3.3.1)	Polyclonal (4.34)
Isoenzyme (3.1.16)	Potency (4.35)
Katal (3.3.2)	Precision (1.16)
Label (4.28)	Primary Calibration Material (1.19.2.1)
Ligand (4.29)	Principal Assigned Value (1.19.5)

ALPHABETICAL LISTING OF TERMS
(continued)

Procedure (1.17)	Sample (1.24)
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Pseudo-First order Reaction (3.2.2)	Sequential Two Point (3.4.3.1)
Pseudo-Zero Order Reaction (3.2.1)	Serum (2.1.8)
Qualitative Analysis (1.4.1)	Specific System Value (1.19.6)
Quantitative Analysis (1.4.2)	Specificity (Analytical) (1.25)
Radioimmunoassay (4.36.2)	Specificity (Immunological) (4.39)
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Radioreceptor Assay (4.36.3)	Substrate (3.1.20)
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Receptor (4.37)	Two Point (3.4.2)
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Reference Method (1.14.2)	Units Used to Express Enzyme Activity(3.3)
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Reproducibility (1.21)	Value (1.19.4)
Result (1.22)	Value, Conventional True (1.28)
Reverse Passive Agglutination (4.5.5)	Value, True (1.27)
Run (1.23)	Variable Time (3.4.2)
Saliva (2.3.4)	