COLLEGE of AMERICAN PATHOLOGISTS
Laboratory Quality Solutions

Glossary

Principles of Analytic Validation of Immunohistochemical Assays: Guideline Update

Glossary of Guideline Terms

Term	Definition for the Purpose of This Guideline
Accuracy	The degree of correctness or true values of a given laboratory result comparing to a gold standard.[1] *Note*: As IHC assays lack gold standards, this guideline will use concordance.
Analyte specific reagent	Antibodies, both polyclonal and monoclonal, or similar reagents which, through specific binding or chemical reaction with substances in a specimen, are intended for use in a diagnostic application for identification and quantification of an individual chemical or ligand in biological specimens.[2,3]
Assay, IHC	The technical components of the immunohistochemical testing process, exclusive of interpretation or reporting.[4]
Biomarker	A physiological analyte that is objectively measured and evaluated as an indicator of normal biological and pathogenic processes or expected pharmacological responses to a specified therapeutic intervention.[5]
Companion diagnostic assay	An in vitro diagnostic device that provides information that is essential for the safe and effective use of a corresponding therapeutic product. For FDA-approved therapeutics, the use of a companion diagnostic is typically stipulated in the labelling of both.[6,7]
Complementary diagnostic assay	Assays that identify a biomarker-defined subset of patients that typically respond to a drug and aid risk/benefit assessments for individual patients, but that are not pre-requisites for receiving the drug.[6]
Concordance, overall	Also known as percent agreement, is a measure used for comparison of the results of the new test to those obtained using a non-gold standard reference assay (or an “imperfect standard”).[8]
Concordance, negative	The proportion of “negative” samples in which the index test is negative.[8]
Concordance, positive	The proportion of “positive” samples in which the index test is positive.[8]
False negative	A negative test result for a patient or specimen that is known or subsequently proved positive for the condition or constituent in question.[4]
False positive	A positive test result for a patient or specimen that is known or subsequently proved to be negative for the condition or constituent in question.[4]
FDA-approved assay	Assays that are approved for marketing under the FDA premarket approval process for new devices, requiring demonstration of safety and effectiveness of Class III devices.[2,9]
FDA-cleared assay	Assays that are cleared for marketing under the FDA 510(k) review, and such clearance is reserved for devices that are substantially equivalent to those already on the market for which there is a predicate IHC device.[2] *Note*: Less stringent than FDA premarket approval.
FDA Class I IHC: Diagnostic markers, Nonpredictive markers	IHCs being used as adjuncts to conventional histopathologic diagnostic examination and with readily available internal and external control materials. These IHC results are evaluated and incorporated into the diagnostic interpretation by the pathologist.[3,8]
FDA Class II IHC	IHCs intended for the detection and/or measurement of certain target analytes by immunological techniques in order to provide prognostic and predictive data that are not directly confirmed by routine histopathologic internal and external control specimens. These IHCs provide the pathologist with diagnostic information that is ordinarily reported as independent diagnostic information to the ordering clinician.[3,8]

FDA Class III IHC	IHCs that do not meet the criteria for class I or II. Manufacturers of these IHCs must submit valid scientific evidence to support the new intended uses (FDA approval/clearance). IHCs identifying new, clinically significant target analytes in tissue specimens that cannot be confirmed by conventional histopathologic examination.[3]
Fit for purpose	An assay that has been successfully validated for the intended use, combining both laboratory and clinical definitions, and factoring in the disease, diagnostic assay, and where applicable, the drug.[10,11]
Laboratory developed test or assay (LDT)	A type of in vitro diagnostic test that is designed, manufactured and used within a single laboratory according to the laboratory's own procedures. *Note*: LDTs may derive from any of the following scenarios: (1) A testing laboratory develops and validates an IHC assay from first principles using separately purchased, commercially available components (aka "de novo LDT"); (2) A testing laboratory adds/subtracts/modifies any manufacturer specified preanalytical, analytical, or postanalytical component/aspect of a commercially available, regulatory agency–approved IHC assay/in vitro diagnostic device, or uses it for a purpose other than intended by the manufacturer.[1,5]
Laboratory Modified Test	FDA-cleared/approved assays with modification. Modified assays are considered LDTs. *Note*: Relevant modifications include but are not limited to changes to the manufacturer's supplied ingredients, instrumentation or procedure, as well as change of specimen type, specimen preanalytics, or stated purpose of the test, its approved test population, or any claims related to interpretation of the results.[2]
Optimization	The process by which the laboratory serially tests and modifies components of the assay to maximize the signal to noise ratio prior to validating the assay for specific clinical purposes.
Predictive value, negative (NPV)	Probability that a person who has tested negative does not have the biomarker present.[12]
Predictive value, positive (PPV)	Probability that a person who has tested positive actually has the biomarker present.[12]
Predictive marker	Biomarker used to identify individuals who are more likely than similar individuals without the biomarker to experience a favorable or unfavorable effect from a (targeted) therapy.[7,13] *Note*: Predicts responsiveness to a specific treatment among cases of the same diagnosis; independent of other histopathologic findings.
Prognostic marker	Biomarker used to identify likelihood of a clinical event, disease recurrence, or progression in patients, regardless of the treatment.[4,5,7]
Purpose	Intended use at the time the test was developed.[11] *Note*: See Fit-for-Purpose.
Readout	The determination of the intensity, extent, quality, and cellular localization of immunohistochemical signal.
Repeatability	Within run reproducibility.[2]
Reproducibility	Extent of agreement among results obtained by replicate testing of specimen sets between laboratories, testing platforms or readers.[2,8] *Note*: Similar to precision, for qualitative testing.
Revalidation	A process to assess a previously validated test's accuracy and reliability in detecting the marker of interest when there has been a change in test conditions, such as methods, reagents, instrumentation, fixation, specimen types, purpose.[1]
Robustness	Assay reproducibility in the face of changes in various test conditions, such as relevant range of preanalytical conditions, instruments, operators.[12]
Sensitivity, diagnostic	The proportion of those with the target condition (as defined by a reference standard) who test positive with a candidate test.[5] *Note*: As most IHC assays lack a gold standard, this guideline uses concordance.
Sensitivity, analytical	The ability to obtain positive results in concordance with positive results obtained by the reference method.[2]
Specificity, analytical	The ability to obtain negative results in concordance with negative results obtained by the reference method.[2]
Specificity, diagnostic	The proportion of those without the target condition (as defined by a reference standard) who test negative with a candidate assay.[5]

	Note: As most IHC assays lack a gold standard, this guideline uses concordance.
Validation	A process to establish that the performance of a test, tool, or instrument is acceptable for its intended purpose. Validation establishes the performance characteristics of an assay as well as the assay limitations.[5]
Validation, analytical (technical)	The process used to confirm with objective evidence that a laboratory-developed or modified FDA-cleared/approved test method or instrument system delivers reliable results for the intended application.[7,13]
Validation, indirect clinical	The process used to determine that an assay delivers reliable results as compared to a designated previously *clinically* validated reference assay. The comparator assay may or may not be FDA-approved, but it must be qualified/validated in a prospective clinical trial, with established link to clinical outcome.[5]
Verification, analytical	A process by which a laboratory determines that an unmodified FDA-cleared/ approved test performs according to the specifications set forth by the manufacturer when used as directed.[13]

Abbreviations: FDA, Food and Drug Administration; IHC, immunohistochemistry

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