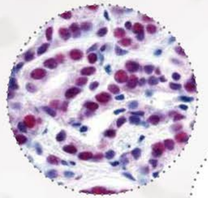
Antibody Selection Tips for PTMs

Preparation:

From an antibody production point of view, the differences between modified proteins can be quite small. Peptide design and immunogen quality are critical to the generation of a specific immune response to ensure to the production of high-quality antibodies.



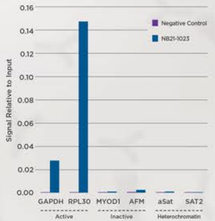
Immunohistochemistry of rabbit anti-HDAC-1 antibody

Production:

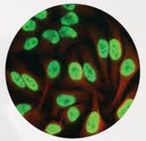
Antibodies against PTMs are generated using a short, specific region of the protein, largely eliminating the issue of specificity seen with antibodies generated using large constructs as immunogens. However, it is critical that the antibody be tested against established positive and negative controls to ensure specificity for the modification. Polyclonal antibodies can be immunodepleted during production if the sample contains antibodies that recognize other PTMs.

Validation:

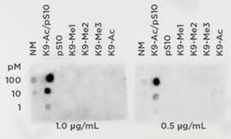
Dot blot assays and ELISAs can be used to assess both antibody specificity and sensitivity. Keep in mind that, in addition to being specific for the required modification, the antibody must be validated for the application of choice using appropriate positive and negative controls.



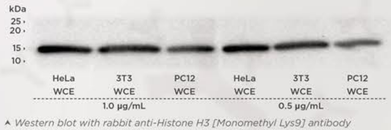
Chromatin immunoprecipitaion with rabbit anti-Histone H3 K4/me3 antibody.



Immunofluorescence of Histone H3 [Lys36ac] (green), DAPI (blue), and alpha-tubulin (red)



Dot blot with rabbit anti-Histone H3 [ac Lys9/phospho Ser10] antibody



Western blot with rabbit anti-Histone H3 [Monomethyl Lys9] antibody

Validated • Reproducible • Multifunctional • Accurate • Dependable

Sponsored by:

ROCKLAND

Mastering Post-Translational Modifications

Cellular regulation beyond gene expression

Post-translational modifications (PTMs) play a key role in dynamic cellular processes, regulating gene expression, protein activity, localization, and degradation, as well as protein interaction. Modification-specific antibodies offer a versatile tool for the characterization of post-translational modifications. Learn how you can choose the best high-affinity, high-specificity antibody for your PTM detection needs.

Custom publishing from:

TheScientist
EXPLORING LIFE, INSPIRING INNOVATION

Common PTMs & Their Functions

Phosphorylation:

Protein phosphorylation is controlled by kinases and phosphatases, and plays a significant role in a wide range of cellular processes, including cell growth and proliferation, metabolism, physiological regulation, and cell signaling.

SUMOylation:

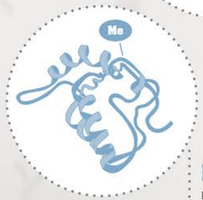
Sumoylation involves the addition of small ubiquitin-like modifiers (SUMOs) that enhance stability or modulate the subcellular compartmentalization of proteins. It has been implicated in various cellular processes, such as nuclear transport, signal transduction, stress response, and cell cycle progression.

Glycosylation:

Attachment of glycans to proteins is critical for protein folding, stability, targeting, and binding. Five types of glycosylation are observed: N- and O-linked glycosylation, C-linked mannosylation, glypiation, and phospho-serine glycosylation.

Acetylation:

Acetylation, or the addition of an acetyl group at lysine residues, is a major post-translational modification for histones, regulating gene expression and metabolism.



Methylation:

Protein methylation is a reversible process by which methyl groups are added to arginine or lysine residues, mediated by peptidylarginine or lysine methyltransferases.

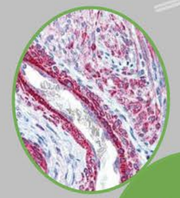
Ubiquitination:

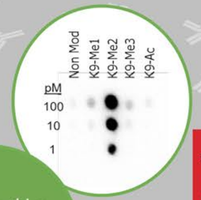
Ubiquitination is an essential cellular process that tags abnormal, foreign, and improperly folded proteins, targeting them for degradation by the 26S proteasome.

Palmitoylation:

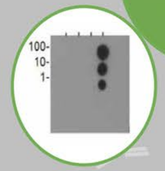
S-Palmitoylation involves the lipid modification of cystine residues with palmitic acid. This modification plays a role in protein localization, stability, subcellular trafficking, and protein-protein interaction.

Cellular regulation beyond gene expression





Find everything you need for your PTM assays in one place.



Highly specific, highly sensitive antibodies that recognize PTMs

Quality control procedures to ensure reproducibility

Validated by researchers at prestigious research institutions

IHC

IP

ChIP

1-800-656-7625

Rockland Antibodies

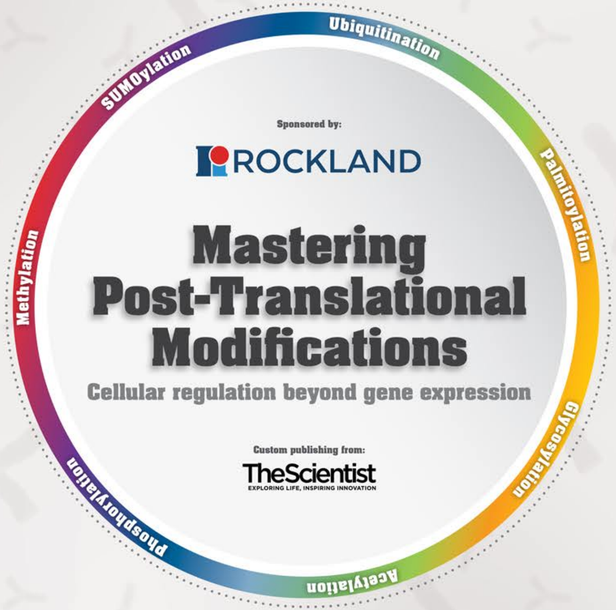
Rockland Immunochemicals, Inc. offers each academic, biopharma, and diagnostic professional thousands of antibodies with the aim of providing the right antibody that is the perfect fit for every occasion. No matter the context—basic research to disease therapy, phosphorylated to methylated and beyond—Rockland embraces the challenge to design, produce, validate, and deliver the absolute best antibodies and life science reagents available in the market today and every day.

Post-translational changes alter the structure of individual proteins, and therefore potentially affect their activity, stability, localization and/or interacting partner molecules. Antibodies are arguably the most prevalent and valuable tool for tracking these changes. Rockland has developed and perfected a process for manufacturing antibodies to detect Post-Translational Modifications (PTM) that has been in use for over a decade. In the process, modification-specific antibodies are prepared using synthetic modified peptides. The trouble is that antibodies recognizing nonphosphorylated forms must be excluded —a skill that Rockland has delicately mastered through years of experience. To ensure the integrity of these sensitive antibodies, Rockland performs quality control testing on every lot to guarantee antibodies function in the intended assays. All work is performed in Rockland's laboratories, located just outside of Philadelphia, PA.

For over fifty years, we at Rockland have assembled an outstanding team of scientists and technicians with a singular dedication to making great antibodies fit for the exacting needs of scientific discovery. From start to finish, we think, innovate, refine, troubleshoot, deliberate, hone, solve, synthesize, purify, conjugate, digest, quantify, qualify, test, package, ship, and guarantee. As we manufacture and validate your antibody, whether selected from our catalog or custom-made, we are keen and intent to deliver reproducible and reliable results in your assay. By ensuring that each step of the process can be certified and validated multiple times, we can achieve our goal to provide accountability and repeatable test results with each antibody we develop. Protect your experiments with Rockland antibodies.

ROCKLAND

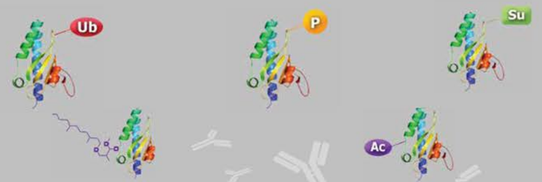
Validated • Reproducible • Multifunctional • Accurate • Dependable



ROCKLAND

Post-Translational Modification Antibodies

At Rockland, scientists have developed proprietary methods for the development of highly specific PTM antibodies that can be used in a wide range of in vitro and in vivo studies of a modified protein, some of which are not easily performed by other approaches, such as mass spectometry.



ELISA

WB

IF

www.rockland.com