

ClearBand HRP Conjugated Goat anti-mouse IgG Antibody

Troubleshooting Guide

500 µL

Cat No: HRPM5

Use this guide for common Western blot and ELISA detection problems with ClearBand HRP Conjugated Goat anti-mouse IgG Antibody. The affinity-purified goat polyclonal secondary antibody is HRP-conjugated, reacts with whole-molecule mouse IgG and mouse immunoglobulin light chains, and is supplied in PBS containing 50% glycerol and 0.05% ProClin 300.

Before You Start - Quick Checks

1. Store the antibody undiluted at -20°C and protect it from prolonged exposure to light.
2. Confirm that the primary antibody is mouse-derived before using this anti-mouse secondary antibody.
3. Do not assume a universal working dilution; the datasheet does not specify one, so optimize the secondary-antibody concentration for the assay system.
4. Use clean, consistent blocking and wash conditions and an HRP-compatible detection substrate.

Problem	Most likely cause	Recommended action
No or very weak signal	The secondary antibody may not match the host species of the primary antibody, or the working concentration may be too low.	Confirm that the primary antibody is mouse-derived, then optimize the secondary-antibody concentration and verify the HRP detection substrate and imaging system.
High background across the membrane or plate	The secondary antibody may be too concentrated, blocking may be insufficient, or washing may be inadequate.	Titrate the secondary antibody to a lower working concentration and optimize blocking and wash steps while keeping the primary-antibody conditions unchanged.
Multiple nonspecific bands	Excess secondary antibody or species cross-reactivity may contribute to nonspecific detection.	Reduce the secondary-antibody concentration and confirm that the sample system does not contain immunoglobulins that can be recognized nonspecifically.
Signal appears with the wrong primary-antibody host	The antibody is directed against mouse IgG and can also recognize mouse immunoglobulin light chains.	Use the secondary antibody only with the intended mouse-primary workflow unless cross-reactivity has been specifically validated.
Signal varies between experiments	Working dilution, incubation, washing, or exposure conditions may not be standardized.	Use the same antibody dilution, incubation time, wash conditions, substrate preparation, and imaging settings across experiments.
Signal decreases after storage	The antibody may have been exposed to unsuitable temperature, repeated handling, or prolonged light.	Keep the antibody undiluted at -20°C and protected from prolonged light exposure; minimize unnecessary warming and handling.
Strong signal but poor signal-to-noise ratio	The antibody/substrate combination may be too sensitive for the current exposure or secondary concentration.	Reduce secondary-antibody concentration and/or shorten imaging exposure while maintaining adequate washing.
Unexpected signal in non-mouse systems	The datasheet notes minimal cross-reaction with rabbit, human and rat immunoglobulins, but cross-reaction with other species may occur.	Interpret unexpected species signals cautiously and validate cross-reactivity experimentally for the sample system being used.

Still having trouble? Recheck primary-antibody host species, secondary-antibody titration, blocking/washing, HRP substrate preparation, exposure settings, and -20°C light-protected storage before changing the primary antibody.