

Certificate of Analysis

Catch and Release[®] v2.0
(Reversible Immunoprecipitation System)
Catalog # 17-500
Lot # DAM1478860

Kit Components

Antibody Capture Affinity Ligand, Catalog # 20-216. One vial containing **60 µg** Antibody Capture Affinity Ligand in **600 µL** PBS containing 2 mM PMSF and 10% glycerol. Store at 4°C.

Catch and Release[®] Wash Buffer, 10X, Catalog # 20-210. One vial containing **15 mL** of 10X buffer, pH 7.4 containing the following detergents: 10% NP-40, 2.5% deoxycholic acid and 150 mM imidazole. Store at 4°C. **Note:** If crystallization occurs when buffer is stored at 4°C, warm to room temperature and vortex briefly before use.

Catch and Release[®] Non-denaturing Elution Buffer, 4X, Catalog # 20-209. One vial containing **10 mL** of 4X PBS-based IP Elution Buffer. Store at 4°C.

Catch and Release[®] Denaturing Elution Buffer, 1X, Catalog # 20-284. One vial containing **4 mL** of 1X Tris-based IP Elution Buffer. Add β-mercaptoethanol (BME) to a final concentration of 5% v/v immediately before use. Store at 4°C.

Catch and Release[®] v2.0 Spin Columns, Catalog # 20-285. **50 columns** containing 0.5ml of prepacked IP capture resin. Store at 4°C

Catch and Release[®] Capture Tubes, 100, 2mL reservoir tubes.

FOR RESEARCH USE ONLY
NOT FOR USE IN HUMANS

Kit Description

Quantity: 50 Immunoprecipitations per kit.

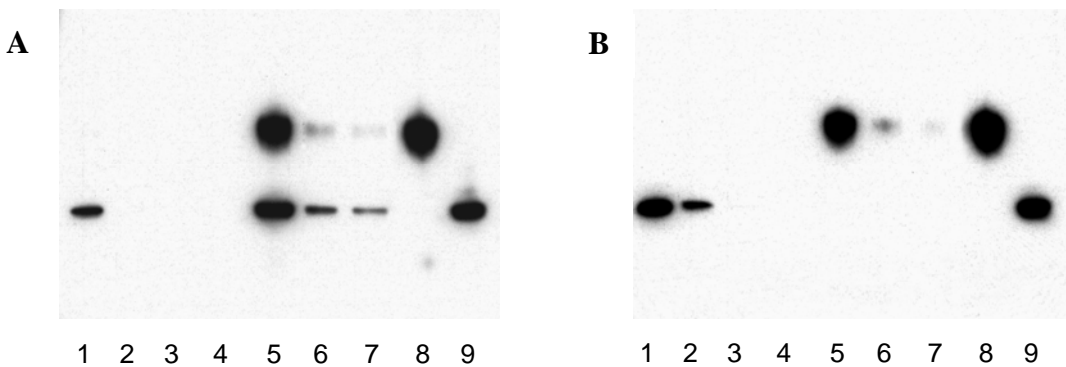
Storage and Stability: All components to be stored at 4°C except the Catch and Release[®] Capture Tubes which are stored at room temperature. Components are stable for 6 months from date of shipment.

Use: This kit allows for quick and reproducible immunoprecipitation (IP) by using a spin column. The system is more reproducible than regular IP's, which are problematic with regards to washing the protein A/G agarose without disrupting the agarose bed. The binding of the antibody/antigen complex in Catch and Release[®] is reversible, and elution of the immune complex can occur with native or denaturing buffers. The system has been tested successfully with rabbit, mouse, sheep and goat antibodies. IP using human IgG1-4 should be suitable. IP using chicken antibodies or human IgA, IgD, IgE or IgM is not recommended with this kit. Please read the enclosed product manual before use.

Application References:

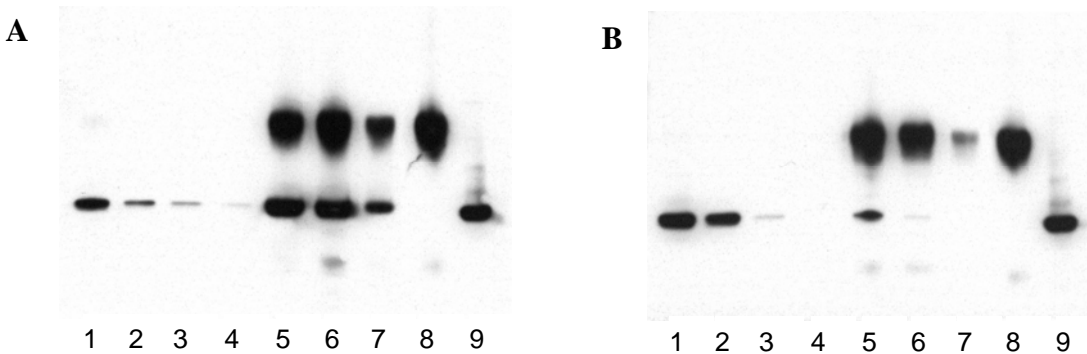
1. Li, *et al.*, *J. Biol. Chem.*, **280**: 6036-6046, 2005.
2. Sastri, *et al.*, *PNAS* **102**: 349-354, 2005.
3. Digicaylioglu, *et al.*, *PNAS* **101**: 9855-9860, 2004.
4. Song, *et al.*, *Mol. Endocrinol.*, **18**: 70-85, 2004.

Quality Control Testing



Catch and Release[®] with Denaturing Elution Buffer:

Catch and Release[®] columns and protocol were used with the Denaturing Elution Buffer to immunoprecipitate cdk2. HeLa nuclear extract was mixed with A. Anti-cdk2 (Catalog # 06-505) or B. normal, rabbit IgG as a negative control for 1 hour at room temperature. Samples from each fraction were run on an SDS-PAGE gel and immunoblotted. The upper band is the heavy chain of IgG and the lower band is cdk2. Lane 1: flow through; Lane 2: wash 1; Lane 3: wash 2; Lane 4: wash 3; Lane 5: elution 1; Lane 6: elution 2; Lane 7: elution 3; Lane 8: Anti-cdk2; Lane 9: HeLa nuclear extract.



Catch and Release[®] with Non-denaturing Elution Buffer:

Catch and Release[®] columns and protocol were used with the Non-denaturing Elution Buffer to immunoprecipitate cdk2. HeLa nuclear extract was mixed with A. Anti-cdk2 (Catalog # 06-505) or B. normal, rabbit IgG as a negative control for 1 hour at room temperature. Samples from each fraction were run on an SDS-PAGE gel and immunoblotted. The upper band is the heavy chain of IgG and the lower band is cdk2. Lane 1: flow through; Lane 2: wash 1; Lane 3: wash 2; Lane 4: wash 3; Lane 5: elution 1; Lane 6: elution 2; Lane 7: elution 3; Lane 8: Anti-cdk2; Lane 9: HeLa nuclear extract.

Unless otherwise stated in our catalog or other company documentation accompanying the product(s), our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans or animals.