

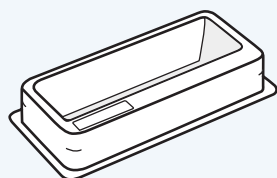
ELISA Workflow Guide

OVERVIEW

Enzyme Linked Immunosorbent Assay (ELISA) is accurate, highly sensitive, and specific for identifying protein species. ELISA microplates enable a common laboratory procedure to be carried out on multiple samples simultaneously. Popular formats include 96-well microplates, 384-well microplates, and 8-well strips.

This guide provides an overview of the tools you'll need at each stage of the ELISA workflow, as well as a few tips for choosing the optimal microplate for your particular assay. Corning is a leading manufacturer of high quality, high performance ELISA microplates and 1 x 8 Corning® Stripwell™ microplates for a wide range of laboratory assays. Corning also carries an extensive variety of accessories that can be used as part of the ELISA workflow, including a full line of buffers, pipettors, pipet tips, and tubes to meet unique assay needs.

Reservoirs



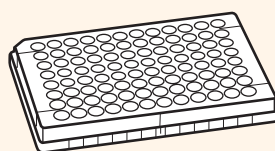
Costar® Reagent Reservoirs

- ▶ Use with multi-channel pipettors to transfer samples, buffers, or reagents into ELISA microplates

Axygen® Multi-channel Reservoirs

- ▶ Single and multiple well formats for manual and automated platforms
- ▶ Multi-channel reservoirs allow for separation of reagents during ELISA preparation

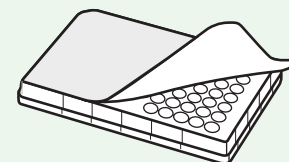
Microplates



Corning Microplates

- ▶ Available in clear, black, or white polystyrene to suit various detection methods: absorbance, fluorescence, or luminescence
- ▶ Medium or High Binding surfaces most commonly used for biochemical assays based on size of target molecule
- ▶ Additional surfaces are available to support other assay types

Microplate Seals

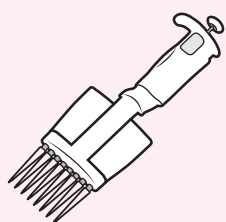


Axygen ELISA Microplate Sealing Films

- ▶ Used during repetitive incubation steps during ELISAs to reduce reagent evaporation, which can cause an “edge effect”

Corning Aluminum Microplate Sealing Tape

- ▶ Utilize during incubation steps to protect light-sensitive samples or reagents for direct and sandwich ELISA



Axygen Multi-channel pipettors

- ▶ Fully autoclavable and UV-resistant for sample protection against contaminations
- ▶ Retracking shafts for perfect tip loading and easier tip ejection
- ▶ Universal fit with all common brands of pipet tips
- ▶ Volume setting protected with a locking system

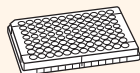
BASIC WORKFLOW

Process Step

Products Used

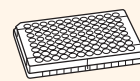
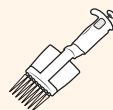
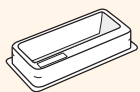
Select the Correct Microplate and Surface for the Application

- ▶ Well format (96-well, 384-well, 8-well strips)
- ▶ Surface chemistry (medium bind, high bind, or other surface type)
- ▶ Microplate color (clear, black, white)



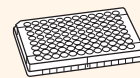
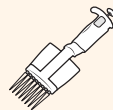
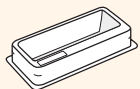
Coat

- ▶ Coat the microplate surface with solution of either antigen or antibody of interest
- ▶ Incubate
- ▶ Wash to remove unbound material



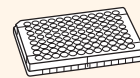
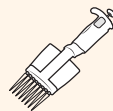
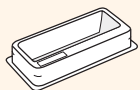
Block (if required)

- ▶ Add protein-based solution to block unbound sites on microplate surface (BSA, Casein)
- ▶ Incubate
- ▶ Wash to remove excess blocking solution (if required)



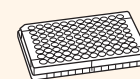
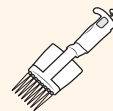
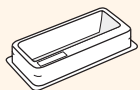
Add Detection Substrate Solution

- ▶ Enzyme, fluorophore-conjugated Ab or direct binding
- ▶ Aluminum sealing tape (ideal for fluorescent reactions)
- ▶ Incubate at the appropriate temperature for the detection method used



Read Signal

- ▶ Add stop solution (if required)
- ▶ Measure produced signal via absorbance, fluorescence, or luminescence



PRODUCTS

For a full list of all microplates, reservoirs, multi-channel pipettors, and microplate seals, visit www.corning.com/lifesciences.

Microplates

Color

Clear – Best suited for absorbance detection.

Black – Low background fluorescence and low fluorescent cross-talk. The black colorant reduces background, as well as light scattering, resulting in higher signal-to-noise ratios.

White – Enhances luminescence signal-to-noise ratio by reflecting light back into the range of the detector.

Surface Chemistry

Medium Binding Surface

- ▶ Hydrophobic
- ▶ Ideal for large, hydrophobic biomolecules (>20 kD)
- ▶ Binding capacity: ~200 ng IgG/cm²

High Binding Surface

- ▶ Hydrophobic and ionic (negatively charged)
- ▶ Ideal for positively charged biomolecules (>10 kD)
- ▶ Binding capacity: ~500 ng IgG/cm²

Corning® ELISA Microplates

Cat. No.	Type	Color	Surface	Qty/Pk	Qty/Cs
9017	96-well, flat-bottom	Clear	Medium binding	25	100
9018	96-well, flat-bottom	Clear	High binding	25	100
3912	96-well, flat-bottom	White	Medium binding	25	100
3922	96-well, flat-bottom	White	High binding	25	100
3915	96-well, flat-bottom	Black	Medium binding	25	100
3925	96-well, flat-bottom	Black	High binding	25	100
2593	Stripwell™ 96-well, flat-bottom	Clear	Medium binding	25	100
2592	Stripwell 96-well, flat-bottom	Clear	High binding	25	100
3923	Stripwell 96-well, flat-bottom	White	High binding	25	100
3924	Stripwell 96-well, flat-bottom	Black	High binding	25	100
3700	384-well, flat-bottom	Clear	High binding	25	100
3702	384-well, flat-bottom	Clear	Not treated	25	100
3576	384-well, flat-bottom	White	High binding	10	50
3572	384-well, flat-bottom	White	Not treated	10	50
3577	384-well, flat-bottom	Black	High binding	10	50
3573	384-well, flat-bottom	Black	Not treated	10	50

Reagent Reservoirs

- ▶ Costar® reagent reservoirs are manufactured from modified polystyrene, are sterile, and disposable.
- ▶ Axygen® single- and multi-channel reservoirs are automation compatible. The multi-channel versions allow for separation of reagents, and are available with up to 12-channels.

Cat. No.	Brand	Channel	Volume	Color
4870	Costar	Single	50 mL	White
4872	Costar	Single	100 mL	White
RES-SW96-HP	Axygen	Single	240 mL	Clear
RES-MW4-HP	Axygen	Four	280 mL (70 mL/channel)	Clear

Sealing Film/Tape

- ▶ Axygen® sealing film is polyester-based with uniformly applied acrylic adhesive to reduce edge effect for sensitive ELISAs, and is suitable for short-term storage/incubation of samples and reagents.
- ▶ Corning® aluminum sealing tape is ideal for use with light-sensitive samples and reagents.

Cat. No.	Brand	Description	Dimension (mm)	Working Temp.	Qty/Pk	Qty/Cs
PCR-SP	Axygen	Polyester, 80 µm sealing film	146 x 79.6	104°C	100	500
6570	Corning	Aluminum sealing tape (96-well)	117.5 x 79.4	-80°C to 150°C	100	100
6569	Corning	Aluminum sealing tape (384-well)	137.2 x 82.5	-80°C to 150°C	100	100

Multi-channel Pipettors and Tips

- ▶ Axygen Axyjet® Pro multi-channel pipettors come in a variety of configurations, are fully autoclavable, and UV resistant.
- ▶ Color-coded pipetting push buttons enable easy size identification.
- ▶ Axygen MultiRack pipet tips are free of detectable RNase, DNase, DNA, and pyrogens. Maxymum Recovery® surface ensures minimum liquid loss and aerosol filters reduce cross-contamination.

Cat. No.	Brand	Description	Pipettor Volume Range	Recommended Pipet Tip Cat. No.	Maximum Tip Volume	Tip Description
AP-8-10-P	Axygen	8-channel	0.5-10 µL	MRF-10XT-L-R-S	10 µL	Extended length, filtered, Maxymum Recovery surface, racked, sterile
AP-8-200-P	Axygen	8-channel	20-200 µL	MRF-200NX-L-R-S	200 µL	Extended length, filtered, Maxymum Recovery surface, racked, sterile

ELISA Technical Documents

Five ELISA Application Notes are available at www.corning.com/lifesciences.

- ▶ Immobilization Principles – Selecting the Surface for ELISA Assays (Corning Lit. Code CLS-DD-AN-454)
- ▶ Optimizing the Immobilization of Protein and Other Biomolecules for ELISA Assays (Corning Lit. Code CLS-DD-AN-455)
- ▶ Effective Blocking Procedures in ELISA Assays (Corning Lit. Code CLS-DD-AN-456)
- ▶ Optimizing the Separation Step on 96-well Microplates for ELISA Assays (Corning Lit. Code CLS-DD-AN-457)
- ▶ Selecting the Detection System – Colorimetric, Fluorescent, Luminescent Methods for ELISA Assays (Corning Lit. Code CLS-DD-AN-458)

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