ELISA Workflow Guide

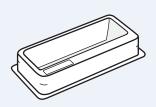


OVERVIEW

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, pipets, tips, and tubes to meet unique assay needs.

Reservoirs



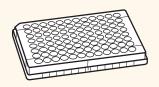
Costar® Reagent Reservoirs

 Use with multi-channel pipets 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/Falcon® 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 lightsensitive samples or reagents for direct and sandwich ELISA

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

Microplates

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

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 signalto-noise ratios.

White – Enhances luminescence signal-tonoise 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 lgG/cm²

Corning® 96-well ELISA Microplates

| Cat. No. | Туре | Color | Surface | Qty/Pk | Qty/Cs | |
|----------|-------------|-------|----------------|--------|--------|--|
| 3591 | Flat-bottom | Clear | Medium binding | 1 | 50 | |
| 3590 | Flat-bottom | Clear | High binding | 1 | 100 | |
| 9017 | Flat-bottom | Clear | Medium binding | 25 | 100 | |
| 9018 | Flat-bottom | Clear | High binding | 25 | 100 | |
| 3912 | Flat-bottom | White | Medium binding | 25 | 100 | |
| 3922 | Flat-bottom | White | High binding | 25 | 100 | |
| 3915 | Flat-bottom | Black | Medium binding | 25 | 100 | |
| 3925 | Flat-bottom | Black | High binding | 25 | 100 | |
| | | | | | | |

Falcon® 96-well ELISA Microplates

| Cat. No. | Туре | Color | Surface | Qty/Pk | Qty/Cs |
|----------|-----------------------|-------|-------------|--------|--------|
| 351172 | Flat-bottom, with lid | Clear | Not treated | 1 | 50 |
| 351190 | Round-bottom | Clear | Not treated | 25 | 100 |

Corning Stripwell™ 96-well ELISA Microplates

| Cat. No. | Туре | Color | Surface | Qty/Pk | Qty/Cs |
|----------|-------------|-------|----------------|--------|--------|
| 2593 | Flat-bottom | Clear | Medium binding | 25 | 100 |
| 2592 | Flat-bottom | Clear | High binding | 25 | 100 |
| 3923 | Flat-bottom | White | High binding | 25 | 100 |
| 3924 | Flat-bottom | Black | High binding | 25 | 100 |

Corning 384-well ELISA Microplates

| Cat. No. | Туре | Color | Surface | Qty/Pk | Qty/Cs |
|----------|-------------|-------|--------------|--------|--------|
| 3700 | Flat-bottom | Clear | High binding | 25 | 100 |
| 3702 | Flat-bottom | Clear | Not treated | 25 | 100 |
| 3576 | Flat-bottom | White | High binding | 10 | 50 |
| 3572 | Flat-bottom | White | Not treated | 10 | 50 |
| 3577 | Flat-bottom | Black | High binding | 10 | 50 |
| 3573 | Flat-bottom | Black | Not treated | 10 | 50 |
| | | | | | |

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Costar® Reagent Reservoirs

- Manufactured from modified polystyrene
- Sterile
- Disposable
- > 50 mL or 100 mL volume

| Cat No. | Volume | Color | Qty/Pk | Qty/Cs |
|---------|--------|-------|--------|--------|
| 4870 | 50 mL | White | 5 | 200 |
| 4872 | 100 mL | White | 5 | 200 |

Axygen® Single- and Multi-channel Reservoirs

- Complies with ANSI/SLAS footprint for automation
- Multi-channels allow separation of multiple assays and buffers during ELISA
- Ranges from 1- to 12-channel reservoirs

| Cat No. | Channel | Color | Volume/Channel | Total Volume | Qty/Pk | Qty/Cs |
|-------------|---------|-------|----------------|---------------------|--------|--------|
| RES-SW96-HP | Single | Clear | 240 mL | 240 mL | 25 | 25 |
| RES-MW4-HP | Four | Clear | 70 mL | 280 mL | 25 | 25 |

Axygen ELISA Microplate Sealing Film

- Polyester-based with uniformly applied acrylic adhesive that reduces the edge effect of sensitive ELISA assays
- Suitable for short-term storage and incubation
- Utilized for incubation of ELISA assays and buffers

| Cat No. | Material | Thickness | Dimension (mm) | Working Temp. | Qty/Pk | Qty/Cs |
|---------|-----------|-----------|----------------|---------------|--------|--------|
| PCR-SP | Polyester | 80 μm | 146 x 79.6 | 104°C | 100 | 500 |

Corning® Aluminum Microplate Sealing Tape

For light sensitive samples and reagents

| Cat No. | Material | Microplate | Qty/Pk | Qty/Cs |
|---------|----------|------------|--------|--------|
| 6570 | Aluminum | 96-well | 100 | 100 |
| 6569 | Aluminum | 384-well | 100 | 100 |

ELISA Technical Documents

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

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

CORNING

Corning Incorporated Life Sciences

836 North St.
Building 300, Suite 3401
Tewksbury, MA 01876
t 800.492.1110
t 978.442.2200
f 978.442.2476
www.corning.com/lifesciences

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