

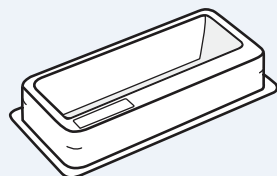
# ELISA Workflow Guide

## 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



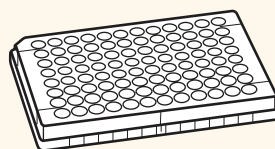
#### Costar® Reagent Reservoirs

- ▶ 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

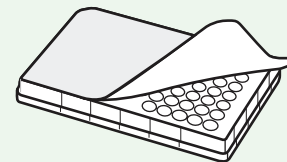
### Microplates



#### 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 light-sensitive samples or reagents for direct and sandwich ELISA

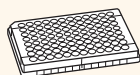
# BASIC WORKFLOW

## Process Step

## Products Used

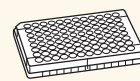
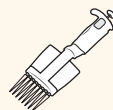
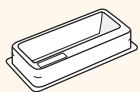
### Select the Correct Microplate and Surface for the Application

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



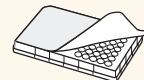
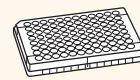
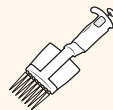
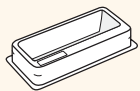
### Coat

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



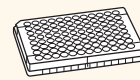
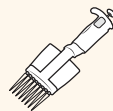
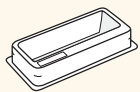
### Block (if required)

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



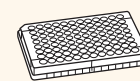
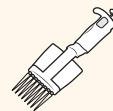
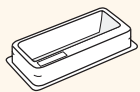
### Add Detection Substrate Solution

- ▶ 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](http://www.corning.com/lifesciences)

### Color

**Clear** – Best suited for absorbance detection

**Black** – Low background fluorescence and low fluorescent cross-talk. The black colorant used in Corning® microplates 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/cm<sup>2</sup>

#### High Binding Surface

- ▶ Hydrophobic and ionic (negatively charged)
- ▶ Ideal for positively charged biomolecules (>10 kD)
- ▶ Binding capacity: ~500 ng IgG/cm<sup>2</sup>

### Corning 96-well ELISA Microplates

| Fisher Scientific Cat. No. | Corning Cat. No. | Type        | Color | Surface        | Qty/Pk | Qty/Cs |
|----------------------------|------------------|-------------|-------|----------------|--------|--------|
| 10337891                   | 3591             | Flat-bottom | Clear | Medium binding | 1      | 50     |
| 10544522                   | 3590             | Flat-bottom | Clear | High binding   | 1      | 100    |
| 10596091                   | 9017             | Flat-bottom | Clear | Medium binding | 25     | 100    |
| 10293532                   | 9018             | Flat-bottom | Clear | High binding   | 25     | 100    |
| 10002551                   | 3912             | Flat-bottom | White | Medium binding | 25     | 100    |
| 10679141                   | 3922             | Flat-bottom | White | High binding   | 25     | 100    |
| 10022561                   | 3915             | Flat-bottom | Black | Medium binding | 25     | 100    |
| 10133811                   | 3925             | Flat-bottom | Black | High binding   | 25     | 100    |

### Falcon® 96-well ELISA Microplates

| Fisher Scientific Cat. No. | Corning Cat. No. | Type                  | Color | Surface     | Qty/Pk | Qty/Cs |
|----------------------------|------------------|-----------------------|-------|-------------|--------|--------|
| 10748692                   | 351172           | Flat-bottom, with lid | Clear | Not treated | 1      | 50     |
| 10693652                   | 351190           | Round-bottom          | Clear | Not treated | 25     | 100    |

### Corning Stripwell™ 96-well ELISA Microplates

| Fisher Scientific Cat. No. | Corning Cat. No. | Type        | Color | Surface        | Qty/Pk | Qty/Cs |
|----------------------------|------------------|-------------|-------|----------------|--------|--------|
| 10367221                   | 2593             | Flat-bottom | Clear | Medium binding | 25     | 100    |
| 10517691                   | 2592             | Flat-bottom | Clear | High binding   | 25     | 100    |
| 10296621                   | 3923             | Flat-bottom | White | High binding   | 25     | 100    |
| 10419331                   | 3924             | Flat-bottom | Black | High binding   | 25     | 100    |

### Corning 384-well ELISA Microplates

| Fisher Scientific Cat. No. | Corning Cat. No. | Type        | Color | Surface      | Qty/Pk | Qty/Cs |
|----------------------------|------------------|-------------|-------|--------------|--------|--------|
| 10134431                   | 3700             | Flat-bottom | Clear | High binding | 25     | 100    |
| 10481242                   | 3702             | Flat-bottom | Clear | Not treated  | 25     | 100    |
| 10359854                   | 3576             | Flat-bottom | White | High binding | 10     | 50     |
| 10109632                   | 3572             | Flat-bottom | White | Not treated  | 10     | 50     |
| 10713767                   | 3577             | Flat-bottom | Black | High binding | 10     | 50     |
| 10745303                   | 3573             | Flat-bottom | Black | Not treated  | 10     | 50     |

## Costar® Reagent Reservoirs

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

| Fisher Scientific Cat. No. | Corning Cat. No. | Volume | Color | Qty/Pk | Qty/Cs |
|----------------------------|------------------|--------|-------|--------|--------|
| 10320551                   | 4870             | 50 mL  | White | 5      | 200    |
| 10712541                   | 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

| Fisher Scientific Cat. No. | Corning Cat. No. | Channel | Color | Volume/Channel | Total Volume | Qty/Pk | Qty/Cs |
|----------------------------|------------------|---------|-------|----------------|--------------|--------|--------|
| 11370275                   | RES-SW96-HP      | Single  | Clear | 240 mL         | 240 mL       | 25     | 25     |
| 12537867                   | 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

| Fisher Scientific Cat. No. | Corning Cat. No. | Material  | Thickness | Dimension (mm) | Working Temp. | Qty/Pk | Qty/Cs |
|----------------------------|------------------|-----------|-----------|----------------|---------------|--------|--------|
| 12772796                   | PCR-SP           | Polyester | 80 µm     | 146 x 79.6     | 104°C         | 100    | 500    |

## Corning® Aluminum Microplate Sealing Tape

For light sensitive samples and reagents

| Fisher Scientific Cat. No. | Corning Cat. No. | Material | Microplate | Qty/Pk | Qty/Cs |
|----------------------------|------------------|----------|------------|--------|--------|
| 10629141                   | 6570             | Aluminum | 96-well    | 100    | 100    |
| 10651272                   | 6569             | Aluminum | 384-well   | 100    | 100    |

## ELISA Technical Documents

Five ELISA Application Notes are available at [www.corning.com/lifesciences](http://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)

For more specific information on claims, visit [www.corning.com/certificates](http://www.corning.com/certificates).

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