



LAMPIRE BIOLOGICAL LABORATORIES

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ELISA Testing Form

Investigator:	Phone: (if questions)
	Email:
Company/Institution:	

See Page 2 for a summary of Lampire's Standard ELISA format. Any changes to the standard protocol may result in additional charges.

If any "Other" box is checked, this constitutes a custom ELISA to which a custom 77##### part number below is assigned.

ELISA - Titer Screen	Testing - Immunoassay	7700560
ELISA - Inhibition	Testing - Immunoassay	7700561
ELISA - Recovery	Testing - Immunoassay	7700562

Services	Selections
1. Plate Coating	<input type="checkbox"/> 1 µg/well <input type="checkbox"/> Other _____
2. Blocking Agent	<input type="checkbox"/> BSA <input type="checkbox"/> Other _____
3. Initial Serum Dilution	<input type="checkbox"/> 1:100 tittered 1:10 in duplicate <input type="checkbox"/> Other _____
4. Incubation	<input type="checkbox"/> 37 °C for 1 hour <input type="checkbox"/> Other _____
5. Washing	<input type="checkbox"/> PBS/Tween 20 <input type="checkbox"/> Other _____
6. Secondary Ab	<input type="checkbox"/> HRP <input type="checkbox"/> Other _____
7. Substrate	<input type="checkbox"/> ABTS <input type="checkbox"/> Other _____
8. Additional Requests	



ELISA Testing Form

Summary of Lampire's Standard ELISA format.

Plate Coating:

- Sufficient plates are coated at the start of the project to run all bleeds originally agreed upon in the scope of work
- Plates are coated at 1ug/well or 100ug/10mL.
- Plates are blocked with BSA unless the injection antigen is conjugated to BSA, then a different blocking agent will be used
- All columns are coated except column 12 of the 96 well plate. This column will serve as the plate blank in testing

Plate Running:

- An internal positive control is run on each day of ELISA testing and statistically analyzed to ensure the positive control falls within the historical data trend
- Initial serum dilution is 1:100 and the material is tittered 1:10 across the plate in duplicate rows. Antibody is added to all columns except column 11 and column 12. Column 11 will have all reagents except antibody added to it and used as a reagent blank. Column 12 is used as a plate blank.
- All incubations are at 37°C for 1 hour
- Plates are washed using PBS with Tween 20
- Secondary antibody conjugated to HRP is used that has been cross adsorbed against serum proteins. New lots of secondary antibody are tested against the previous lot to ensure that reproducible results are maintained from test to test when a new lot of material is received. Dilutions of the secondary antibody are matched to produce similar results
- ABTS substrate is added to the plates and stopped after the appropriate time with ABTS solution

Analysis:

- Plates are read at 405nm wavelength and interpreted with SoftMax Software. The Curve Fit option for Fixed Weight value. Titers of the animals are determined as 50% of the highest OD obtained for the animal and is reported out by the software based on the OD values and curve generated.