Protein A ELISA Kit (9777-1)

User Guide

For the detection of NGL-Impact® A HipH

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Abbreviations

C	Celsius
dH_2O	Distilled water
F	Fahrenheit
HPLC	High-performance liquid chromatography
HRP	Horseradish peroxidase
µL	1000000 microliter
µm	Micrometer
mm	Millimeter
ng/mg	Nanograms to milligrams
ng/mL	Nanograms per milliliter
NGL	Next generation ligand
Nm	Nanometers
PBS	Phosphate buffered saline
pH	Is a measure of how acidic/basic water is
PLC	Programmable logic controller
ppm	Parts per million
TMB	Tetramethylbenzidine

1. Overview of ELISA

The Protein A ELISA Kit (9777-1) from Repligen provides accurate and precise quantitation of residual Protein A. The kit has been developed towards reliability, ease-of-use and sensitivity to quantitate low concentrations of Protein A in therapeutic protein samples.

Testing for residual Protein A occurs in several different phases of development and commercial manufacturing that may include:

- Process development: leaching characteristics of the resin under specific conditions
- Manufacturing: eluted samples taken throughout several points in the purification process
- Finish product release: document process containment levels and lot-to-lot consistency

The Protein A ELISA Kit includes the NGL-Impact® A HipH standard, one of several Protein A ligands used in monoclonal antibody affinity chromatography. Repligen also offers Protein A ELISA Kits for the MabSelect SuRe™ (part number 9333-1) and the rProtein A (part number 9000-1) ligands.

The capture plate in this kit is coated with anti-Protein A antibodies. NGL-Impact® A HipH Standards and Test Samples are diluted with sample diluent (Reagent A) and incubated with the immobilized antibodies. Captured NGL-Impact® A HipH is then quantitated by the addition of Biotinylated anti-Protein A probe (Reagent C). The high biotin density of the probe allows high dynamic binding capacity of Streptavidin Peroxidase conjugate (Reagent D). The final detection step involves adding Tetramethylbenzidine, TMB (Reagent E), to give a colorimetric reaction. Color intensity is proportional to the amount of NGL-Impact® A HipH present in the sample.

1.1 Important points regarding assay sensitivity

1. The units of the assay results are expressed as nanograms per milliliter (ng/mL) of NGL-Impact® A HipH.
2. The limit of quantitation, defined as 10-fold the standard deviation of a blank sample, is typically 0.1 ng/mL.
3. For the Dilute and Go method, the lowest concentration in the starting sample that can be measured is typically 0.4 ng/mL. With the recommended starting concentration of 0.5 mg/mL antibody, this corresponds to 0.8 ppm.
4. For the Boil and Boost method the lowest concentration in the starting sample that can be measured is typically 1.0 ng/mL. With the recommended starting concentration of 10 mg/mL, this corresponds to 0.01 ppm.
5. Assay characterization recommendations are available in Repligen Technical Notes. Please contact Customer Service for a copy or go to www.repligen.com.

Table 1. Reagents provided

Reagent	Description	Volume	Storage
Reagent A	Sample diluent (5X) concentrate	20 mL	2 - 8° C
Reagent B	NGL-Impact® A HipH Standard solution, contains 1.0 mg/mL ligand in sterile water	200 µL	2 - 8° C
Reagent C	Rabbit anti-Protein A: Biotin probe, contains 0.02% sodium azide	200 µL	2 - 8° C
Reagent D	Streptavidin-HRP (Horseradish Peroxidase) conjugate	200 µL	2 - 8° C
Reagent E	TMB Peroxidase substrate, contains 3, 3', 5, 5'-tetramethylbenzidine in buffer	20 mL	2 - 8° C
PBS packs	Final volume of each pack when reconstituted is 1 L	2 packs	Ambient
ELISA Plate	96-well microtiter plate coated with anti-Protein A antibodies (packed with desiccants)	Dried Plate	2 - 8° C

Note: *Reagents are specific to the kit lot and should be discarded once all plate strips have been consumed.*

Table 2. Equipment, reagents, and supplies not provided with kit

Equipment	Reagents and Supplies
1 L graduated cylinder	dH2O or HPLC-Grade water (preferred)
Micro-pipettors and 12-channel pipettor	1.5 mL Microcentrifuge tubes
ELISA plate reader with wavelength capability at 450 nm	15 mL and 50 mL plastic centrifuge tubes
Timer	Polysorbate 20
Vortex mixer	Reagent reservoirs
Micro-centrifuge	5 mL and 10 mL Serological pipettes
Water bath	Phosphoric acid
Plate sealers	Filter (0.22 µm) and 1 L bottle

2. Sample preparation

Select appropriate sample preparation method (Section 3.2 of User Guide) and prepare samples.

Method	Description
A – Buffer Exchange	Buffer-exchange into PBS (by dialysis or spin column) then dilute to 0.5 mg/mL of antibody in PBS prior to test sample dilution prep (Section 3.4 of User Guide)
B – Dilute and Go	Dilute in PBS 0.1% Polysorbate 20 at least ten-fold, to 0.5 mg/mL of antibody then perform test sample dilution prep (Section 3.4 of User Guide)
C – Boil and Boost	1) Dilute samples to ≤ 15 mg/mL of antibody, if necessary, in neutral buffer 2) adjust samples to 0.1% Polysorbate 20. Samples 3) boil for 5 minutes and centrifuged prior to test sample dilution prep (Section 3.4)

Reagent preparation

1. Make 1X sample diluent: 4.0 mL Reagent A + 16 mL dH_20.
2. Prepare PBS solution: reconstitute 1 PBS pack in 1L water and filter through 0.2 µm.
3. Make PBS Polysorbate 20: 700 mL PBS + 700 µL Polysorbate 20; filter through 0.2 µm.

Microtiter plate setup

1. Design experiment and set up the 12 microtiter well strips as needed.
2. Make sure all wells are placed correctly placed and level and wash 3x with HPLC or distilled grade water.

Antibody sample preparation

1. For each sample: 200 µL Reagent A + 550 µL dH_20 and vortex.
2. Add 250 µL of sample (1:4 dilution), vortex, and let stand 5 – 10 minutes at room temperature.

Note: *For Methods A and B, antibody concentration must be ≤ 0.5 mg/mL.*

Protein A standard curve

1. Make Protein A Standard solution:
   Tube 1: 10 µL Reagent B + 990 µL 1X sample diluent
   Tube 2: 10 µL Tube 1 + 990 µL 1X sample diluent
   Tube 3: 25 µL Tube 2 + 975 µL 1X sample diluent

Note: *Tube 3 is the 2.5 ng/mL standard solution.*

2. Vortex and let stand 10 minutes at room temperature before sequential serial dilutions.

Preparation of 2-fold serial dilutions of antibody sample

These instructions describe preparation of a 7-point standard curve in triplicate. The standard curve samples and antibody samples are used starting in wells H1 – H3 and D4 – D6 respectively.

1. Add 100 µL of 1X sample diluent solution into all wells that will contain serially diluted antibody samples or serially diluted Protein A Standard. Do not add sample diluent to wells that will contain initial aliquots of antibody sample or Protein A Standard.
2. Transfer 200 µL of the 2.5 ng/mL standard solution (Tube 3) into wells H1 – H3.
3. Transfer 200 µL of the prepared antibody sample into wells D4 – D6.
4. Make 2-fold serial dilutions of the Protein A Standard and antibody sample simultaneously by transferring 100 µL up to next row.
5. Discard 100 µL of solution from the final wells containing serial diluted standard or sample, leaving 100 µL in each well.

ELISA testing

1. After serial dilutions, seal the plate with film and incubate for 30 minutes at room temperature.
2. Allow TMB substrate (Reagent E) to come to room temperature (protect from light).
3. Wash plate with PBS Polysorbate 4 times.
4. Prepare probe: 70 µL Reagent C + 12 mL PBS Polysorbate.
5. Add 100 µL diluted probe to each well except wells A1 – A3 (substrate blanks).
6. Seal the place and incubate 30 minutes at room temperature,
7. Prepare conjugate: 12 µL Reagent D + 12 mL PBS Polysorbate.
8. Add 100 µL conjugate solution to each well except wells A1 – A3.
9. Incubate 30 minutes at room temperature.
10. After 30 minutes, wash 2 times with PBS Polysorbate and then 2 times with PBS only.
11. Add 100 µL TMB substrate to each well and incubate 4 minutes.
12. Stop reaction with 100 µL of 1N Phosphoric acid.
13. Read the plate at 450 nm.

3. Guide to standard preparation and assay

3.1 Pre-Assay reagent preparation

All ELISA Kit components

Allow all kit components to equilibrate to *room temperature.

1X sample diluent

Dilute 4.0 mL of Reagent A (5X sample diluent) in 16 mL of purified water in a 50 mL plastic centrifuge tube. Vortex for 5 – 20 seconds or invert 10 – 15 times for thorough mixing. If required, the 1X sample diluent is stable for 2 weeks at *room temperature.

PBS solution

Dissolve the contents of one PBS pack in 800 mL of dH_20 to a final volume of 1L. Mix well. Filter PBS solution through a 0.22 µm filter.

PBS Polysorbate 20 wash solution
Pour 700 mL of the PBS solution (prepared and filtered per instructions above) into a 1 L graduated cylinder. Add 700 µL of Polysorbate 20. Mix well. Save the remaining 300 mL PBS solution for the final ELISA wash. Filter PBS Polysorbate solution through a 0.22 µm filter.

TMB substrate solution
For a full-plate assay, use the whole bottle of TMB. For half-plate assays, aliquot 8 mL of TMB into a 15 mL conical centrifuge tube and wrap the tube with aluminum foil to limit light exposure. Return bottle to the 2 – 8° C refrigerator.

Test samples — Allow all test samples to equilibrate to *room temperature. Prepare a 10% Polysorbate 20 solution only if using the Boil and Boost method.

***Note:** An ideal room temperature range of 65 - 77° F (18 - 25° C) is important for optimum assay performance.*

3.2 Sample preparation methods

Multiple sample preparation methods for the NGL-Impact® A HipH ELISA assay have been developed to allow end users to select the method most appropriate for individual samples. Table 3 describes a representative preparation method.

Table 3. Preparation method overview (with starting concentrations)

Method	Input antibody concentration	Description
A – Buffer Exchange	N/A	Buffer-exchange samples into PBS (by dialysis or spin column) then dilute antibody to 0.5 mg/mL in PBS prior to test sample dilution prep (Section 3.4)
B – Dilute and Go	≥ 5.0 mg/mL	Dilute samples in PBS 0.1% Polysorbate 20 at least ten-fold, to 0.5 mg/mL antibody, before performing test sample dilution prep (Section 3.4)
C – Boil and Boost	≤ 15 mg/mL	Dilute antibody to ≤ 15 mg/mL if necessary, in neutral buffer. Adjust sample composition to 0.1% Polysorbate 20. Boil samples for 5 minutes and centrifuge prior to test sample dilution prep (Section 3.4)

Table 4. Method attributes

	A: Buffer Exchange	B: Dilute and Go	C: Boil and Boost
Assay completion < 2 hours	✓	✓	✓
Reduced sample preparation steps		✓	
Enhanced limit of quantitation			✓
High starting sample concentration			✓

Method A: Buffer exchange
Prior to running the assay, samples must be buffer-exchanged into PBS (0.01 M phosphate buffer, 0.15 M sodium chloride, 0.003 M potassium chloride, pH 7.2 - 7.4) and diluted to a protein concentration of ≤ 0.5 mg/mL. Dialysis or a desalting column may be used.

***Note:** The PBS packs provided in the kit are not intended for this buffer exchange. They are to be reconstituted and used as directed in the ELISA protocol.*

Method B: Dilute and Go
This method dilutes interfering substances to support common process buffers such as 100 mM Citrate, Glycine, and Acetic buffers neutralized with Tris-base. Prior to running the assay, dilute Protein A-purified antibody samples with starting concentrations greater than 5.0 mg/mL directly into phosphate buffered saline (PBS) with 0.1% Polysorbate 20 to reach a final concentration of 0.5 mg/mL. For best performance, characterize assay performance with process-specific buffers and proteins.

Note: *No buffer exchange is required when the dilution step is performed. If sample concentration is less than 5.0 mg/mL, the Dilute and Go method (Method B) is not recommended. Instead, the user should proceed with buffer exchange (Method A).*

Method C: Boil and Boost
This method is designed for high analyte concentration samples. Compatibility with common process buffers such as 100 mM Citrate and Acetate neutralized with Tris-base at antibody concentrations up to 15 mg/mL has been demonstrated. Characterize assay performance with process-specific buffers and proteins.

Note: *Buffers with glycine or with > 0.2% Polysorbates can negatively impact the limit of quantitation. Samples containing glycine or high concentrations of surfactants should be buffer exchanged into PBS prior to running the boil and boost method.*

Add at least 0.5 mL of each sample to a 1.5 mL centrifuge tube (the assay procedure will require 0.25 mL.) Polysorbate 20 should be added to each sample to a final concentration of 0.1% (can use the 10% Polysorbate solution made for the boiling method). Create a pin hole in the cap of each centrifuge tube and boil for 5 minutes in a water bath. After cooling the samples, centrifuge the tubes at 13,000 x g for 5 minutes. Boiling causes disassociation and precipitation of antibodies or Fc-fusions from Protein A molecules. Transfer the supernatant to a new tube (optional). The supernatant will be used when preparing sample dilutions in the assay procedure.

3.3 Standard preparation

Table 5. Concentrated standard solution preparation

Tube	Protein A Standard	1X sample diluent
1	10 µL of Reagent B	990 µL
2	10 µL of Tube 1	990 µL
3	25 µL of Tube 2	975 µL

1. When Reagent B is completely thawed, vortex to mix. If reagent remains on the sides or cap of the tube, briefly spin in a microcentrifuge.
2. Label three 1.5 mL microcentrifuge tubes as Tube 1, Tube 2, and Tube 3. Prepare the target concentration standard solution (2.5 ng/mL NGL-Impact® A HipH, Tube 3) by diluting Reagent B with 1X sample diluent per in the table below. (Vortex each tube thoroughly between dilutions).
3. Keep Tube 3 (2.5 ng/mL NGL-Impact® A HipH Protein A Standard) for use later.

3.4 Test sample dilution preparation

1. After test samples have been prepared and are at the appropriate starting concentration (Table 3), label a microcentrifuge tube for each test sample. Add 200 µL of 5X sample diluent (Reagent A) to each. Add 550 µL of dH_2O to each tube. Vortex for 5 – 10 seconds.

2. Equilibrate all samples to room temperature before diluting. Add 250 µL of each test sample to the labeled tubes. Vortex for 5 – 10 seconds. These are the first 1:4 starting sample dilutions. Reserve the tubes for future use.
3. Incubate all test samples and the 2.5 ng/mL standard dilution for 10 minutes at room temperature before pipetting into the assay plate.
4. During the 10-minute incubation, wash the plate three times. Fill the wells with dH_2O by using a wash bottle or automated plate-washing system. Remove the liquid from the plate and repeat. After the third wash, dry the plate by inverting it on clean paper towels and tapping gently.

3.5 Plate set-up

Note: *The following pipetting and dilution instructions describe a single sample assay, as shown in Table 6. Analogous steps should be taken when processing multiple samples. Alternatively, users may choose to prepare standards and samples in a dilution plate and transfer to the assay plate.*

1. Using a 12-channel pipettor, add 100 µL of 1X sample diluent into columns 1 – 3 rows B – G and columns 4 - 6 rows A – C.
2. Transfer 200 µL of 2.5 ng/mL NGL-Impact® A HipH Standard solution (Tube 3) into wells H1 – H3.
3. Transfer 200 µL of 1:4 Antibody sample dilution into wells D4 – D6
4. Make 2-fold serial dilutions of the NGL-Impact® A HipH Standard and antibody samples by transferring 100 µL from each set of triplicate wells into the well directly above them. Mix thoroughly by pipetting 5 times.

Note: *In a single sample assay format, the same tips can be used for each row.*

5. After making the last NGL-Impact® A HipH Standard serial dilution in wells C1 – C3, remove 100 µL and discard. Also discard 100 µL from the final antibody sample dilution in wells A4 – A6.

Table 6. Representative plate set-up for one antibody sample

	1	2	3	4	5	6	7	8	9	10	11	12
A	Plate blank			1:32								
B	0 ng/mL			1:16								
C	0.078 ng/mL			1:8								
D	0.157 ng/mL			Sample #1, 1:4								
E	0.313 ng/mL											
F	0.625 ng/mL											
G	1.25 ng/mL											
H	2.5 ng/mL											

3.6 ELISA procedure

1. After the NGL-Impact® A HipH Standards and Antibody sample dilutions have been prepared, seal the plate with film and incubate at room temperature for 30 minutes.
2. After incubation, remove all liquid from the wells. Using a wash bottle or automated plate-washing system, wash the plate with PBS-Polysorbate 20 solution. Remove the liquid and dry thoroughly by inverting the plate on clean paper towels and tapping gently. Repeat the wash and dry cycle three additional times for a total of four washes.

3. Briefly vortex the Reagent C vial. If reagent material remains on the sides or cap of the tube, briefly spin in a micro-centrifuge. Prepare the Rabbit anti-Protein A Biotin probe solution. For a full plate assay, prepare 12 mL by combining 70 µL of Reagent C with 12 mL of prepared PBS Polysorbate 20 in a 15 mL conical centrifuge tube. For a half-plate assay, prepare 6 mL by combining 35 µL of Reagent C with 6 mL PBS Polysorbate 20 in a 15 mL conical centrifuge tube. Mix solution thoroughly.
4. Using a 12-channel pipettor, add 100 µL of the diluted Reagent C probe solution to each well containing a test sample or standard. Leave wells A1 – A3 (Plate blanks) empty.
5. Seal the plate with film and incubate at room temperature for 30 minutes. After incubation, wash the wells four times with PBS Polysorbate 20 and remove the liquid. Dry thoroughly by inverting the plate on clean paper towels and tapping gently.
6. Briefly vortex the Reagent D vial. If reagent material remains on the sides or the cap of the tube, briefly spin in a micro-centrifuge. For a full-plate assay, prepare 12 mL of Streptavidin horseradish Peroxidase conjugate solution by combining 12 µL of Reagent D with 12 mL of prepared PBS Polysorbate 20 in a 15 mL conical centrifuge tube. For a half-plate assay, prepare 6mL by combining 6 µL of Reagent D with 6 mL PBS Polysorbate 20 in a 15 mL conical centrifuge tube. Mix solution thoroughly.
7. Add 100 µL of diluted Reagent D conjugate solution to each well containing test sample or Standard. Leave wells A1 – A3 (Plate blanks) empty.
8. Seal the plate with film and incubate at room temperature for 30 minutes.
9. After incubation, discard the conjugate solution from the plate. Wash the wells twice with PBS Polysorbate 20. Wash twice more but with PBS only. After each wash, discard the liquid by inverting the plate on clean paper towels and tapping gently.

Note: *Before proceeding with the next step, make sure the TMB solution is at room temperature 65 - 77°F (18 - 25°C). If the lab is too warm, move the assay to a cooler location for the development step.*

10. Using a multi-channel pipettor, add 100 µL of the TMB substrate to each of the wells, **including** A1 - A3 (Plate blanks).
11. Incubate plate for 4 minutes. Stop reaction by adding 100 µL of 1N phosphoric acid to each well, including A1 – A3, in the same order of pipetting used for the TMB substrate solution.

Note: *Other strong acids typically used as stop solutions in ELISA may be substituted for 1N phosphoric acid. If bubbles are present in the wells, agitate slightly before reading.*

12. Read the plate at 450 nm within 20 minutes of acid quench.

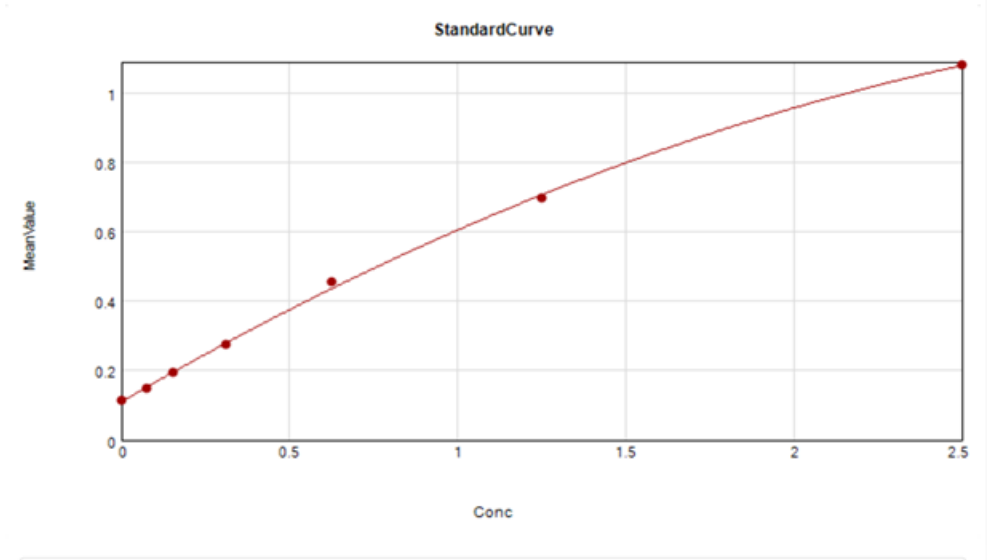
4. Calculation of results

1. Calculate the mean absorbance value for the plate blank wells (A1 – A3) and subtract from all remaining wells on the plate (including the 0 ng/mL standard curve). Determine the mean absorbance value for each standard concentration and all test samples.

Note: *Standard Curve calculation should be based on the standards present on the same plate. Other curve fits may be used as deemed appropriate.*

2. Calculate the Standard Curve:
   Plot each Standard Curve concentration (ng/mL Protein A) on the x-axis and the corresponding mean absorbance value on the y-axis. Using Quadratic, 4-parameter logistical fit or 5-parameter logistical fit, calculate the best-fitting line through the points of the standard curve (Figure 1 and Figure2).

Figure 1. Standard Curve Quadratic Fit



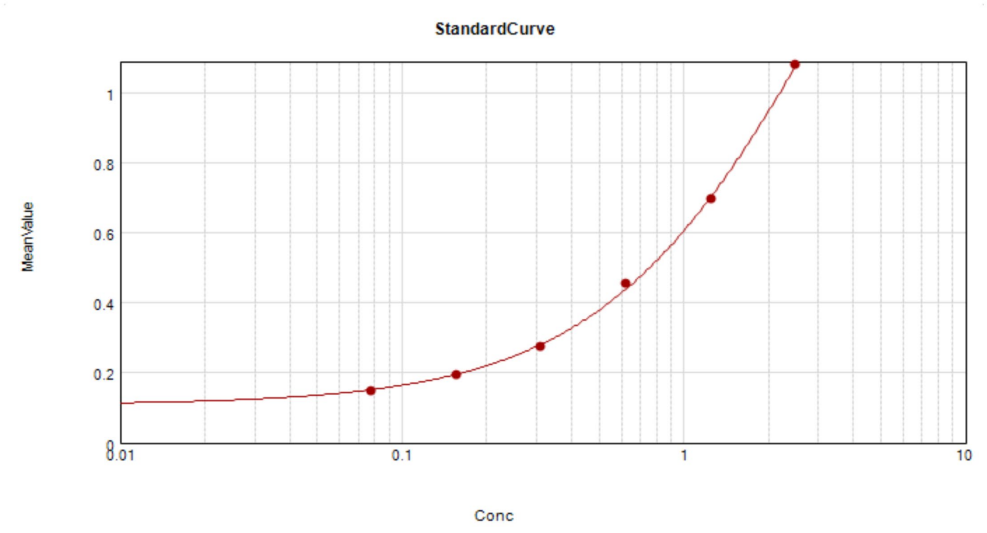
● Std (Standards: MeanValue vs Concentration) Weighting: Fixed

Curve Fit Results ▲

Curve Fit : Quadratic $y = A + Bx + Cx^2$

	Parameter	Estimated Value	Std. Error	Confidence Interval
Std R^2 = 0.999	A	0.111	0.007	[0.090, 0.131]
	B	0.566	0.021	[0.509, 0.624]
	C	-0.072	0.008	[-0.094, -0.049]

Figure 2. Standard Curve 4-Parameter Logistical Fit



● Std (Standards: MeanValue vs Concentration) Weighting: Fixed

Curve Fit Results ▲

Curve Fit : 4-Parameter Logistic $y = D + \frac{A - D}{1 + \left(\frac{x}{C}\right)^B}$

	Parameter	Estimated Value	Std. Error	Confidence Interval
Std R^2 = 0.999 EC50 = 3.580	A	0.109	0.010	[0.076, 0.143]
	B	1.044	0.093	[0.747, 1.342]
	C	3.580	1.393	[-0.852, 8.013]
	D	2.491	0.546	[0.754, 4.228]

The regression line can be used to determine the NGL-Impact® A HipH concentration [PA] for the samples.

$$[PA] \times Sample\ Dilution = C\ (ng/mL)$$

To determine the ng/mg (ppm) of Protein A in each sample well, use the following formula:

$$\frac{ng}{mg} = ppm = \frac{mean\ concentration\ (ng/mL)}{\frac{mg}{mL} of\ antibody\ per\ well\ (e.g., 0.125\ mg/mL)}$$

Specificity
Protein A ELISA Kits (9777-1) are supplied with Repligen NGL-Impact® A HipH Standard Solution. For the behavior of the kit with of other variants of Protein A, please contact Repligen Customer Service customerserviceUS@repligen.com.

5. Troubleshooting

Problem: Pipetting enough of required reagent.

Possible cause	Remedy
Splashing of reagent on sides or cap of reagent tube during mixing, shipping, or handling.	Centrifuge tube briefly.

Problem: Inconsistent results between sample dilutions.

Possible cause	Remedy
Antibody was not fully equilibrated in PBS, pH 7.0 – 7.4, before assay. The antibody concentration in the undiluted sample was > 0.5 mg per mL.	Re-dialyze sample in PBS. Ensure pH is 7.0 – 7.4 and re-run assay. Ensure antibody concentration is < 0.5 mg/mL.

Problem: Outliers, where one replicate has an abnormally high or low absorbance value.

Possible cause	Remedy
Small amount of peroxidase conjugate left on the plate before color development. (i.e., wells were not thoroughly washed)	Discard outliers and average duplicates. Ensure thorough washing in any subsequent ELISA testing.

Problem: Color development time to reach 1.0 AU is > 4 – 5 minutes.

Possible cause	Remedy
TMB solution, Reagent E, was not at room temperature before adding to wells. Room temperature too low, or too cool.	Solution can be warmed before adding to wells. Use incubator set at 65 – 77° F (18 – 25° C) for all for all incubations or develop longer than 4 minutes.

Problem: Background signal is > 0.150 OD Units.

Possible cause	Remedy
Color development for TMB substrate was > 4 minutes. Temperature of TMB substrate > 77° F (25° C). Insufficient plate washing.	Start timer immediately after adding TMB substrate to 2.5 ng/mL Standard wells. Store TMB in a location that is between 65 – 77° F (18 – 25° C) until use. Ensure plate was washed 4 times.

Problem: O.D. values consistently high for all samples, or low recovery of Protein A in samples.

Possible cause	Remedy
Buffer component interference.	Buffer exchange sample into neutral buffer or perform a greater fold dilution into neutral buffer (Section 3.2).

6. Additional references

(1) H. Fey and G. Burkhard, (1981) “Measurement of Staphylococcal Protein A and Detection of Protein A-Carrying Staphylococcus Strains by a Competitive ELISA method” J. Immunol. Methods 47: 99-107.

(2) A. Warnes, A. Walkland and J.R. Stephenson, (1986) “Development of an Enzyme-Linked Immunosorbent Assay for Staphylococcal Protein A Produced in Escherichia coli by pUC8-based Plasmids Containing the Staphylococcus aureus Cowan I protein A Gene” J. Immunol. Methods 93:63-70.

(3) M.T. Dertzbaugh, M.C. Flickinger and W.B. Lebherz III, (1985) “An Enzyme Immunoassay for the Detection of Staphylococcal Protein A in Affinity-Purified Products” J. Immunol. Methods 83: 169-177.

(4) J.W. Bloom, M.F. Wong and G. Mitra, (1989) “Detection and Reduction of Protein A Contamination in Immobilized Protein A-Purified Monoclonal Antibody Preparations” J. Immunol. Methods 117: 83-89.

(5) S.M. Knicker, A.T. Profy, (1991) “Immunoassay to Measure Staphylococcal Protein A in the Presence of Murine Immunoglobulins” J. Immunol. Methods 142: 53-59.

(6) Dudley, R.A., P. Edwards, et al. (1985) “Guidelines for immunoassay data processing.” Clin Chem 31(8): 1264-71.

(7) Smith, W.C. and G.S. Sittampalam (1998) “Conceptual and statistical issues in the validation of analytic dilution assays for pharmaceutical applications.” J Biopharm Stat 8(4): 509-32.

7. Safety Data Sheet example

Figure 3. Reagent A – Component of NGL-Impact® A HipH ELISA Kit 9777-1

REPLIGEN

Reagent A

Safety Data Sheet

According To Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules And Regulations

Date of Issue: 08/11/2021 Version: 1.0

SECTION 1: IDENTIFICATION

1.1. Product Identifier

Product Form: Mixture

Product Name: Reagent A

Synonyms: Reagent A for Kits 9000-1, 9222-1, 9333-1, 9444-1, 9777-1, 9888-1.

1.2. Intended Use of the Product

Use of the Substance/Mixture: Kit Component. Detection of specific standards referenced on each kit label. For R&D use only.

1.3. Name, Address, and Telephone of the Responsible Party

Company

Repligen Corporation
41 Seyon Street, Building 1, Suite 100
Waltham, MA 02453
USA
+1 781-250-0111

customerserviceUS@repligen.com

1.4. Emergency Telephone Number

Emergency Number : ChemTel LLC
(800)255-3924 (North America)
+1 (813)248-0585 (International)

SECTION 2: HAZARDS IDENTIFICATION

2.1. Classification of the Substance or Mixture

Skin corrosion/irritation Category 2 H315 - Causes skin irritation

2.2. Label Elements

GHS-US Labeling

Hazard Pictograms (GHS-US) :

GHS07

Signal Word (GHS-US) : Warning

Hazard Statements (GHS-US) : H315 - Causes skin irritation.

Precautionary Statements (GHS-US) : P264 - Wash hands, forearms, and other exposed areas thoroughly after handling.
P280 - Wear protective gloves, protective clothing, and eye protection.
P302+P352 - If on skin: Wash with plenty of water.
P321 - Specific treatment (see section 4 on this SDS).
P332+P313 - If skin irritation occurs: Get medical advice/attention.
P362+P364 - Take off contaminated clothing and wash it before reuse.

2.3. Other Hazards

Exposure may aggravate pre-existing eye, skin, or respiratory conditions.

2.4. Unknown Acute Toxicity (GHS-US)

No data available

SECTION 3: COMPOSITION/INFORMATION ON INGREDIENTS

3.1. Substance

Not applicable

3.2. Mixture

Name	Synonyms	Product Identifier	%	GHS US classification
Sodium acetate	Acetic acid, sodium salt / Acetic acid, sodium salt (1:1) / Sodium acetate, anhydrous / SODIUM ACETATE / Sodium acetate anhydrous	(CAS-No.) 127-09-3	10 - 20	Skin corrosion/irritation Category 2, H315 - Causes skin irritation Combustible Dust

The specific chemical identity and/or exact percentage of composition have been withheld as a trade secret [29 CFR 1910.1200].

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SECTION 4: FIRST AID MEASURES

4.1. Description of First-aid Measures

First-aid Measures General: Never give anything by mouth to an unconscious person. If you feel unwell, seek medical advice (show the label where possible).

First-aid Measures After Inhalation: When symptoms occur: go into open air and ventilate suspected area. Obtain medical attention if breathing difficulty persists.

First-aid Measures After Skin Contact: Remove contaminated clothing. Immediately drench affected area with water for at least 15 minutes. Obtain medical attention if irritation develops or persists.

First-aid Measures After Eye Contact: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Obtain medical attention if irritation develops or persists.

First-aid Measures After Ingestion: Rinse mouth. Do NOT induce vomiting. Obtain medical attention.

4.2. Most Important Symptoms and Effects Both Acute and Delayed

Symptoms/Injuries: Causes skin irritation.

Symptoms/Injuries After Inhalation: Not expected to be a primary route of exposure. Prolonged exposure may cause irritation.

Symptoms/Injuries After Skin Contact: Redness, pain, swelling, itching, burning, dryness, and dermatitis.

Symptoms/Injuries After Eye Contact: Prolonged exposure may cause slight irritation to eyes.

Symptoms/Injuries After Ingestion: Not expected to be a primary route of exposure. Ingestion may cause adverse effects.

Chronic Symptoms: None known.

4.3. Indication of Any Immediate Medical Attention and Special Treatment Needed

If exposed or concerned, get medical advice and attention. If medical advice is needed, have product container or label at hand.

SECTION 5: FIRE-FIGHTING MEASURES

5.1. Extinguishing Media

Suitable Extinguishing Media: Water spray, fog, carbon dioxide (CO_2), alcohol-resistant foam, or dry chemical.

Unsuitable Extinguishing Media: Do not use a heavy water stream. Use of heavy stream of water may spread fire.

5.2. Special Hazards Arising From the Substance or Mixture

Fire Hazard: Not considered flammable but may burn at high temperatures.

Explosion Hazard: Product is not explosive.

Reactivity: Hazardous reactions will not occur under normal conditions.

5.3. Advice for Firefighters

Precautionary Measures Fire: Exercise caution when fighting any chemical fire.

Firefighting Instructions: Use water spray or fog for cooling exposed containers. Remove containers from fire area if this can be done without risk. Do not breathe fumes from fires or vapors from decomposition.

Protection During Firefighting: Do not enter fire area without proper protective equipment, including respiratory protection.

Hazardous Combustion Products: Carbon oxides (CO, CO_2). Sodium oxides. Nitrogen oxides.

Other Information: Exposure to fire may cause containers to rupture/explode.

SECTION 6: ACCIDENTAL RELEASE MEASURES

6.1. Personal Precautions, Protective Equipment and Emergency Procedures

General Measures: Avoid breathing (vapor, mist, spray). Avoid all contact with skin, eyes, or clothing.

6.1.1. For Non-Emergency Personnel

Protective Equipment: Use appropriate personal protective equipment (PPE).

Emergency Procedures: Evacuate unnecessary personnel.

6.1.2. For Emergency Personnel

Protective Equipment: Equip cleanup crew with proper protection.

Emergency Procedures: Ventilate area. Upon arrival at the scene, a first responder is expected to recognize the presence of dangerous goods, protect oneself and the public, secure the area, and call for the assistance of trained personnel as soon as conditions permit.

6.2. Environmental Precautions

Prevent entry to sewers and public waters.

6.3. Methods and Materials for Containment and Cleaning Up

For Containment: Contain any spills with dikes or absorbents to prevent migration and entry into sewers or streams.

Methods for Cleaning Up: Clean up spills immediately and dispose of waste safely. Absorb and/or contain spill with inert material. Transfer spilled material to a suitable container for disposal. Contact competent authorities after a spill.

6.4. Reference to Other Sections

See Section 7 for handling and storage, Section 8 for exposure controls and personal protection and Section 13 for disposal.

Reagent A

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SECTION 7: HANDLING AND STORAGE

7.1. Precautions for Safe Handling

Additional Hazards When Processed: Contains substances that are combustible dusts. If dried, allowed to accumulate, and dispersed in air, may form combustible dust concentrations in air that could ignite and cause an explosion. Take appropriate precautions.

Precautions for Safe Handling: Do not handle until all safety precautions have been read and understood. Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Avoid contact with skin, eyes and clothing. Avoid breathing vapors, mist, spray. Use appropriate personal protective equipment (PPE).

Hygiene Measures: Handle in accordance with good industrial hygiene and safety procedures. Wash contaminated clothing before reuse.

7.2. Conditions for Safe Storage, Including Any Incompatibilities

Technical Measures: Comply with applicable regulations.

Storage Conditions: Keep container closed when not in use. Store in a dry, cool and well-ventilated place. Keep/Store away from direct sunlight, extremely high or low temperatures and incompatible materials.

Incompatible Materials: Strong acids, strong bases, strong oxidizers. Alkalis. Halogenated compounds. Peroxides. Nitrates.

Storage Temperature: 2 – 8 °C

7.3. Specific End Use(s)

Kit Component. Detection of specific standards referenced on each kit label. For R&D use only.

SECTION 8: EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1. Control Parameters

For substances listed in section 3 that are not listed here, there are no established exposure limits from the manufacturer, supplier, importer, or the appropriate advisory agency including: ACGIH (TLV), AIHA (WEEL), NIOSH (REL), or OSHA (PEL).

8.2. Exposure Controls

Appropriate Engineering Controls : Emergency eye wash fountains and safety showers should be available in the immediate vicinity of any potential exposure. Ensure adequate ventilation, especially in confined areas. Ensure all national/local regulations are observed.

Personal Protective Equipment : Gloves. Protective clothing. Protective goggles. Insufficient ventilation: wear respiratory protection.

Materials for Protective Clothing : Chemically resistant materials and fabrics.

Hand Protection : Wear protective gloves.

Eye and Face Protection : Chemical safety goggles.

Skin and Body Protection : Wear suitable protective clothing.

Respiratory Protection : If exposure limits are exceeded or irritation is experienced, approved respiratory protection should be worn. In case of inadequate ventilation, oxygen deficient atmosphere, or where exposure levels are not known wear approved respiratory protection.

Other Information : When using, do not eat, drink or smoke.

SECTION 9: PHYSICAL AND CHEMICAL PROPERTIES

9.1. Information on Basic Physical and Chemical Properties

Physical State	: Liquid
Appearance	: Colorless liquid
Color	: Colorless
Odor	: Strong, vinegar-like
Odor Threshold	: No data available
pH	: 3
Evaporation Rate	: No data available
Melting Point	: No data available
Freezing Point	: No data available
Boiling Point	: No data available
Flash Point	: No data available

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Auto-ignition Temperature	:	No data available
Decomposition Temperature	:	No data available
Flammability (solid, gas)	:	Not applicable
Vapor Pressure	:	No data available
Relative Vapor Density at 20°C	:	No data available
Relative Density	:	No data available
Solubility	:	Water: Soluble
Partition Coefficient: N-Octanol/Water	:	No data available
Viscosity	:	No data available

9.2. Other Information No additional information available

SECTION 10: STABILITY AND REACTIVITY

10.1. Reactivity: Hazardous reactions will not occur under normal conditions.

10.2. Chemical Stability: Stable under recommended handling and storage conditions (see section 7).

10.3. Possibility of Hazardous Reactions: Hazardous polymerization will not occur.

10.4. Conditions to Avoid: Direct sunlight, extremely high or low temperatures, and incompatible materials. Sources of ignition.

10.5. Incompatible Materials: Strong acids, strong bases, strong oxidizers. Alkalis. Halogenated compounds. Peroxides. Nitrates.

10.6. Hazardous Decomposition Products: Thermal decomposition may produce: Carbon oxides (CO, CO_2). Hydrocarbons. Nitrogen oxides. Sodium oxides.

SECTION 11: TOXICOLOGICAL INFORMATION

11.1. Information on Toxicological Effects

Acute Toxicity (Oral): Not classified (Based on available data, the classification criteria are not met)

Acute Toxicity (Dermal): Not classified (Based on available data, the classification criteria are not met)

Acute Toxicity (Inhalation): Not classified (Based on available data, the classification criteria are not met)

Sodium acetate (127-09-3)	
LD50 Oral Rat	3530 mg/kg
LD50 Dermal Rabbit	> 10 g/kg
LC50 Inhalation Rat	> 30 g/m³ (Exposure time: 1 h)

Skin Corrosion/Irritation: Causes skin irritation.

pH: 3

Serious Eye Damage/Irritation: Not classified (Based on available data, the classification criteria are not met)

pH: 3

Respiratory or Skin Sensitization: Not classified (Based on available data, the classification criteria are not met)

Germ Cell Mutagenicity: Not classified (Based on available data, the classification criteria are not met)

Carcinogenicity: Not classified (Based on available data, the classification criteria are not met)

Reproductive Toxicity: Not classified (Based on available data, the classification criteria are not met)

Specific Target Organ Toxicity (Single Exposure): Not classified (Based on available data, the classification criteria are not met)

Specific Target Organ Toxicity (Repeated Exposure): Not classified (Based on available data, the classification criteria are not met)

Aspiration Hazard: Not classified (Based on available data, the classification criteria are not met)

Symptoms/Injuries After Inhalation: Not expected to be a primary route of exposure. Prolonged exposure may cause irritation.

Symptoms/Injuries After Skin Contact: Redness, pain, swelling, itching, burning, dryness, and dermatitis.

Symptoms/Injuries After Eye Contact: Prolonged exposure may cause slight irritation to eyes.

Symptoms/Injuries After Ingestion: Not expected to be a primary route of exposure. Ingestion may cause adverse effects.

Chronic Symptoms: None known.

SECTION 12: ECOLOGICAL INFORMATION

12.1. Toxicity

Ecology - General : Not classified.

Sodium acetate (127-09-3)	
LC50 Fish 1	> 100 mg/l (Exposure time: 96 h - Species: Danio rerio [semi-static])
EC50 - Crustacea [1]	> 1000 mg/l (Exposure time: 48 h - Species: Daphnia magna)

12.2. Persistence and Degradability

Reagent A	
Persistence and Degradability	Not established.

12.3. Bioaccumulative Potential

Reagent A	
Bioaccumulative Potential	Not established.
Sodium acetate (127-09-3)	
BCF Fish 1	< 10

12.4. Mobility in Soil No additional information available

12.5. Other Adverse Effects

Other Information : Avoid release to the environment.

SECTION 13: DISPOSAL CONSIDERATIONS

13.1. Waste Treatment Methods

Waste Disposal Recommendations: Dispose of contents/container in accordance with local, regional, national, and international regulations.

Additional Information: Container may remain hazardous when empty. Continue to observe all precautions.

Ecology - Waste Materials: Avoid release to the environment.

SECTION 14: TRANSPORT INFORMATION

The shipping description(s) stated herein were prepared in accordance with certain assumptions at the time the SDS was authored, and can vary based on a number of variables that may or may not have been known at the time the SDS was issued.

14.1. In Accordance with DOT Not regulated for transport

14.2. In Accordance with IMDG Not regulated for transport

14.3. In Accordance with IATA Not regulated for transport

SECTION 15: REGULATORY INFORMATION

15.1. US Federal Regulations

Reagent A	
SARA Section 311/312 Hazard Classes	Health hazard - Skin corrosion or Irritation

Sodium acetate (127-09-3)
Listed on the United States TSCA (Toxic Substances Control Act) inventory

15.2. US State Regulations Neither this product nor its chemical components appear on any US state lists, or its chemical components are not required to be disclosed.

SECTION 16: OTHER INFORMATION, INCLUDING DATE OF PREPARATION OR LAST REVISION

Date of Preparation or Latest Revision : 08/11/2021

Other Information : This document has been prepared in accordance with the SDS requirements of the OSHA Hazard Communication Standard 29 CFR 1910.1200
The specific chemical identity and/or exact percentage of composition have been withheld as a trade secret [29 CFR 1910.1200].

GHS Full Text Phrases:

Comb. Dust	Combustible Dust
Skin Irrit. 2	Skin corrosion/irritation Category 2
H315	Causes skin irritation

This information is based on our current knowledge and is intended to describe the product for the purposes of health, safety and environmental requirements only. It should not therefore be construed as guaranteeing any specific property of the product.

SDS US (GHS HazCom)

Figure 4. Reagent D – Component of NGL-Impact® A HipH ELISA Kit 9777-1

REPLIGEN

Reagent D

Safety Data Sheet

According To Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules And Regulations

Revision Date: 11/15/2021 Date of Issue: 08/11/2021 Version: 2.0

SECTION 1: IDENTIFICATION

1.1. Product Identifier

Product Form: Mixture

Product Name: Reagent D

Synonyms: Reagent D for Kits 9000-1, 9222-1, 9333-1, 9444-1, 9547-1, 9777-1, 9888-1

1.2. Intended Use of the Product

Use of the Substance/Mixture: Component of ELISA kit used for the detection of specific standards referenced on each kit label. For R&D use only.

1.3. Name, Address, and Telephone of the Responsible Party

Company

Repligen Corporation
41 Seyon Street, Building 1, Suite 100
Waltham, MA 02453
USA
+1 781-250-0111

customerserviceUS@repligen.com

1.4. Emergency Telephone Number

Emergency Number : ChemTel LLC
(800)255-3924 (North America)
+1 (813)248-0585 (International)

SECTION 2: HAZARDS IDENTIFICATION

2.1. Classification of the Substance or Mixture

GHS-US Classification

Skin sensitization, Category 1A	H317
Hazardous to the aquatic environment - Acute Hazard Category 3	H402
Hazardous to the aquatic environment - Chronic Hazard Category 3	H412

2.2. Label Elements

GHS-US Labeling

Hazard Pictograms (GHS-US) :

GHS07

Signal Word (GHS-US) : Warning

Hazard Statements (GHS-US) : H317 - May cause an allergic skin reaction.
H402 - Harmful to aquatic life.
H412 - Harmful to aquatic life with long lasting effects.

Precautionary Statements (GHS-US) : P261 - Avoid breathing mist, spray, vapors.
P272 - Contaminated work clothing must not be allowed out of the workplace.
P273 - Avoid release to the environment.
P280 - Wear eye protection, protective gloves, protective clothing.
P302+P352 - If on skin: Wash with plenty of soap and water.
P321 - Specific treatment (see Section 4 on this SDS).
P333+P313 - If skin irritation or rash occurs: Get medical advice/attention.
P363 - Wash contaminated clothing before reuse.
P501 - Dispose of contents/container in accordance with local, regional, national, and international regulations.

2.3. Other Hazards

Exposure may aggravate pre-existing eye, skin, or respiratory conditions.

SECTION 3: COMPOSITION/INFORMATION ON INGREDIENTS

3.1. Substance

Not applicable

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3.2. Mixture

Name	Synonyms	Product Identifier	%	GHS US classification
1,2,3-Propanetriol	Glycerin / Glycerine / Glycerol / 1,2,3-Trihydroxypropane / GLYCERIN / Propane-1,2,3-triol	(CAS-No.) 56-81-5	25 – 30	Not classified
5-Chloro-2-methyl-3(2H)-isothiazolone, mixture with 2-methyl-3(2H)-isothiazolone	CMI + MIT in mixture 3:1 / Mixture of 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one / Mixture of 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one (3:1) / Mixture of: 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-2H-isothiazol-3-one (3:1) / 5-Chloro-2-methyl-3(2H)-isothiazolone with 2-methyl-3(2H)-isothiazolone / 3(2H)-Isothiazolone, 5-chloro-2-methyl-, mixture with 2-methyl-3(2H)-isothiazolone (3:1) / Methylisothiazolinone and methylchloroisothiazolinone, in combination / Reaction mass of: 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-2H-isothiazol-3-one (3:1) / Kathon CG 5243 and Kathon CG 243 / Mixture of 5-chloro-2-methylisothiazol-3(2H)-one and 2-methylisothiazol-3(2H)-one / Kathon 886 / Reaction mass 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one (3:1) / Mixture of 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-4-isothiazolin-3-one / 2-Methyl-5-chloro-(2H)-isothiazol-3-one with 2-methyl-(2H)-isothiazol-3-one / Mixture of 2-methyl-1,2-thiazol-3(2H)-one and 5-chloro-2-methyl-1,2-thiazol-3(2H)-one / 2-Methyl-1,2-thiazol-3(2H)-one - 5-chloro-2-methyl-1,2-thiazol-3(2H)-one / 3(2H)-Isothiazolone, 5-chloro-2-methyl-, mixture with 2-methyl-3(2H)-isothiazolone / Reaction mass of: 5-chloro-2- methyl-4-isothiazolin-3-one [EC no. 247-500-7]and 2-methyl-2H - isothiazol-3- one [EC no. 220-239-6] (3:1); reaction mass of: 5-chloro-2-methyl-4-isothiazolin-3-one [EC no. 247-500-7]and 2-methyl-4-isothiazolin-3- one [EC no. 220-239-6] (3:1) / Reaction mass of: 5-chloro-2- methyl-4-isothiazolin3-one and 2-methyl-2H-isothiazol-3-one (3:1) / Reaction mass of: 5-chloro-2-methyl-4-isothiazolin3-one and 2-methyl-2H-isothiazol-3-one (3:1) / methylchloroisothiazolinone/methylisothiazolinone mixture / Methylisothiazolinone and methylchloroisothiazolinone / MIXTURE OF METHYLCHLOROISOTHIAZOLINONE AND METHYLISOTHIAZOLINONE (3:1) WITH MAGNESIUM CHLORIDE AND MAGNESIUM NITRATE	(CAS-No.) 55965-84-9	0.002 – < 0.06	Acute Tox. 3 (Oral), H301 Acute Tox. 2 (Dermal), H310 Acute Tox. 4 (Inhalation:dust,mist), H332 Skin Corr. 1B, H314 Eye Dam. 1, H318 Skin Sens. 1A, H317 Aquatic Acute 1, H400 Aquatic Chronic 1, H410

SECTION 4: FIRST AID MEASURES

4.1. Description of First-aid Measures

First-aid Measures General: Never give anything by mouth to an unconscious person. If you feel unwell, seek medical advice (show the label where possible).

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First-aid Measures After Inhalation: When symptoms occur: go into open air and ventilate suspected area. Obtain medical attention if breathing difficulty persists.
First-aid Measures After Skin Contact: Remove contaminated clothing. Wash affected area with soap and water for at least 15 minutes. Obtain medical attention if irritation/rash develops or persists.
First-aid Measures After Eye Contact: Rinse cautiously with water for at least 15 minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Obtain medical attention.
First-aid Measures After Ingestion: Rinse mouth. Do NOT induce vomiting. Obtain medical attention.

4.2. Most Important Symptoms and Effects Both Acute and Delayed

Symptoms/Injuries: Skin sensitization.
Symptoms/Injuries After Inhalation: Prolonged exposure may cause irritation.
Symptoms/Injuries After Skin Contact: May cause an allergic skin reaction.
Symptoms/Injuries After Eye Contact: May cause slight irritation to eyes.
Symptoms/Injuries After Ingestion: Ingestion may cause adverse effects.
Chronic Symptoms: Exposure may produce an allergic reaction.

4.3. Indication of Any Immediate Medical Attention and Special Treatment Needed

If exposed or concerned, get medical advice and attention. If medical advice is needed, have product container or label at hand.

SECTION 5: FIRE-FIGHTING MEASURES

5.1. Extinguishing Media

Suitable Extinguishing Media: Water spray, fog, carbon dioxide (CO_2), alcohol-resistant foam, or dry chemical. Alcohol resistant foams are preferred. General purpose synthetic foams (including AFFF) or protein foams may function, but will be less effective.
Unsuitable Extinguishing Media: Do not use a heavy water stream. Use of heavy stream of water may spread fire.

5.2. Special Hazards Arising From the Substance or Mixture

Fire Hazard: Not considered flammable but may burn at high temperatures.
Explosion Hazard: Product is not explosive.
Reactivity: Hazardous reactions will not occur under normal conditions.

5.3. Advice for Firefighters

Precautionary Measures Fire: Exercise caution when fighting any chemical fire.
Firefighting Instructions: Use water spray or fog for cooling exposed containers. Do not breathe fumes from fires or vapours from decomposition.
Protection During Firefighting: Do not enter fire area without proper protective equipment, including respiratory protection.
Hazardous Combustion Products: Carbon oxides (CO, CO_2). Nitrogen oxides. Hydrogen chloride. Sulfur oxides. Irritating fumes.
Other Information: Do not allow run-off from fire fighting to enter drains or water courses.

SECTION 6: ACCIDENTAL RELEASE MEASURES

6.1. Personal Precautions, Protective Equipment and Emergency Procedures

General Measures: Avoid breathing (vapor, mist, spray). Do not get in eyes, on skin, or on clothing.

6.1.1. For Non-Emergency Personnel

Protective Equipment: Use appropriate personal protective equipment (PPE).
Emergency Procedures: Evacuate unnecessary personnel.

6.1.2. For Emergency Personnel

Protective Equipment: Equip cleanup crew with proper protection.
Emergency Procedures: Upon arrival at the scene, a first responder is expected to recognize the presence of dangerous goods, protect oneself and the public, secure the area, and call for the assistance of trained personnel as soon as conditions permit. Ventilate area.

6.2. Environmental Precautions

Prevent entry to sewers and public waters. Avoid release to the environment.

6.3. Methods and Materials for Containment and Cleaning Up

For Containment: Contain any spills with dikes or absorbents to prevent migration and entry into sewers or streams.
Methods for Cleaning Up: Clean up spills immediately and dispose of waste safely. Absorb and/or contain spill with inert material. Do not take up in combustible material such as: saw dust or cellulosic material. Transfer spilled material to a suitable container for disposal. Contact competent authorities after a spill.

6.4. Reference to Other Sections

See Section 7 for handling and storage, Section 8 for exposure controls and personal protection and Section 13 for disposal considerations.

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SECTION 7: HANDLING AND STORAGE

7.1. Precautions for Safe Handling

Additional Hazards When Processed: None reasonably foreseeable.

Precautions for Safe Handling: Obtain special instructions before use. Do not handle until all safety precautions have been read and understood. Avoid breathing vapors, mist, spray. Avoid prolonged contact with eyes, skin and clothing. Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Use appropriate personal protective equipment (PPE).

Hygiene Measures: Handle in accordance with good industrial hygiene and safety procedures.

7.2. Conditions for Safe Storage, Including Any Incompatibilities

Technical Measures: Comply with applicable regulations.

Storage Conditions: Store in a dry, cool place. Keep container closed when not in use. Containers which are opened should be properly resealed and kept upright to prevent leakage. Keep/Store away from direct sunlight, extremely high or low temperatures and incompatible materials.

Incompatible Materials: Strong acids, strong bases, strong oxidizers. Reducing agents. Amines. Mercaptans. Nucleophils.

Storage Temperature: 2 – 8 °C (35.6 - 46.4 °F)

Special Rules on Packaging: Keep only in original container.

7.3. Specific End Use(s)

Component of ELISA kit used for the detection of specific standards referenced on each kit label. For R&D use only.

SECTION 8: EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1. Control Parameters

For substances listed in section 3 that are not listed here, there are no established exposure limits from the manufacturer, supplier, importer, or the appropriate advisory agency including: ACGIH (TLV), AIHA (WEEL), NIOSH (REL), or OSHA (PEL).

1,2,3-Propanetriol (56-81-5)		
USA OSHA	OSHA PEL (TWA) [1]	15 mg/m³ (mist, total particulate) 5 mg/m³ (mist, respirable fraction)

8.2. Exposure Controls

Appropriate Engineering Controls : Ensure adequate ventilation, especially in confined areas. Ensure all national/local regulations are observed. Suitable eye/body wash equipment should be available in the vicinity of any potential exposure.

Personal Protective Equipment : Gloves. Protective clothing. Protective goggles.

Materials for Protective Clothing : Chemically resistant materials and fabrics.

Hand Protection : Wear protective gloves.

Eye and Face Protection : Chemical safety goggles.

Skin and Body Protection : Wear suitable protective clothing.

Respiratory Protection : If exposure limits are exceeded or irritation is experienced, approved respiratory protection should be worn. In case of inadequate ventilation, oxygen deficient atmosphere, or where exposure levels are not known wear approved respiratory protection.

Environmental Exposure Controls : Do not allow to enter drains or water courses.

Other Information : When using, do not eat, drink or smoke.

SECTION 9: PHYSICAL AND CHEMICAL PROPERTIES

9.1. Information on Basic Physical and Chemical Properties

Physical State : Liquid

Appearance : Clear to Pink

Odor : No data available

Odor Threshold : No data available

pH : No data available

Evaporation Rate : No data available

Melting Point : No data available

Freezing Point : No data available

Boiling Point : No data available

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Flash Point	: No data available
Auto-ignition Temperature	: No data available
Decomposition Temperature	: No data available
Flammability (solid, gas)	: Not applicable
Vapor Pressure	: No data available
Relative Vapor Density at 20°C	: No data available
Relative Density	: No data available
Solubility	: No data available
Partition Coefficient: N-Octanol/Water	: No data available
Viscosity	: No data available

9.2. Other Information

No additional information available

SECTION 10: STABILITY AND REACTIVITY

10.1. Reactivity

Hazardous reactions will not occur under normal conditions.

10.2. Chemical Stability

Stable under recommended handling and storage conditions (see section 7).

10.3. Possibility of Hazardous Reactions

Hazardous polymerization will not occur.

10.4. Conditions to Avoid

Direct sunlight, extremely high or low temperatures, and incompatible materials.

10.5. Incompatible Materials

Strong acids, strong bases, strong oxidizers. Reducing agents. Amines. Mercaptans. Nucleophils.

10.6. Hazardous Decomposition Products

Thermal decomposition may produce: Acrolein. Carbon oxides (CO, CO_2). Nitrogen oxides. Hydrogen chloride. Sulfur oxides.

SECTION 11: TOXICOLOGICAL INFORMATION

11.1. Information on Toxicological Effects

Acute Toxicity (Oral): Not classified (Based on available data, the classification criteria are not met)

Acute Toxicity (Dermal): Not classified (Based on available data, the classification criteria are not met)

Acute Toxicity (Inhalation): Not classified (Based on available data, the classification criteria are not met)

5-Chloro-2-methyl-3(2H)-isothiazolone, mixture with 2-methyl-3(2H)-isothiazolone (55965-84-9)	
LD50 Oral Rat	53 mg/kg
LD50 Dermal Rabbit	87.12 mg/kg
LC50 Inhalation Rat	1.23 mg/l/4h
ATE (Dermal)	87.12 mg/kg body weight
ATE (Vapors)	1.23 mg/l/4h
ATE (Dust/Mist)	1.23 mg/l/4h

1,2,3-Propanetriol (56-81-5)	
LD50 Oral Rat	12600 mg/kg
LD50 Dermal Rabbit	> 10 g/kg

Skin Corrosion/Irritation: Not classified (Based on available data, the classification criteria are not met)

Serious Eye Damage/Irritation: Not classified (Based on available data, the classification criteria are not met)

Respiratory or Skin Sensitization: May cause an allergic skin reaction.

Germ Cell Mutagenicity: Not classified (Based on available data, the classification criteria are not met)

Carcinogenicity: Not classified (Based on available data, the classification criteria are not met)

Reproductive Toxicity: Not classified (Based on available data, the classification criteria are not met)

Specific Target Organ Toxicity (Single Exposure): Not classified (Based on available data, the classification criteria are not met)

Specific Target Organ Toxicity (Repeated Exposure): Not classified (Based on available data, the classification criteria are not met)

Aspiration Hazard: Not classified (Based on available data, the classification criteria are not met)

Symptoms/Injuries After Inhalation: Prolonged exposure may cause irritation.

Symptoms/Injuries After Skin Contact: May cause an allergic skin reaction.

Symptoms/Injuries After Eye Contact: May cause slight irritation to eyes.

Symptoms/Injuries After Ingestion: Ingestion may cause adverse effects.

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Chronic Symptoms: Exposure may produce an allergic reaction.

SECTION 12: ECOLOGICAL INFORMATION

12.1. Toxicity

Ecology - General : Harmful to aquatic life with long lasting effects.

5-Chloro-2-methyl-3(2H)-isothiazolone, mixture with 2-methyl-3(2H)-isothiazolone (55965-84-9)	
LC50 Fish 1	0.09 mg/l
EC50 - Crustacea [1]	0.007 mg/l
ErC50 (Algae)	0.0107 (0.0107 – 0.0535) mg/l
NOEC Chronic Fish	0.02 mg/l
NOEC Chronic Crustacea	0.1 mg/l
NOEC Chronic Algae	0.00049 mg/l

1,2,3-Propanetriol (56-81-5)	
LC50 Fish 1	54000 (51000 – 57000) mg/l (Exposure time: 96 h - Species: Oncorhynchus mykiss [static])

12.2. Persistence and Degradability

Reagent D	
Persistence and Degradability	May cause long-term adverse effects in the environment.

12.3. Bioaccumulative Potential

Reagent D	
Bioaccumulative Potential	Not established.
1,2,3-Propanetriol (56-81-5)	
BCF Fish 1	(no bioaccumulation)
Partition coefficient n-octanol/water (Log Pow)	-1.76

12.4. Mobility in Soil

Reagent D	
Ecology - Soil	Not established.

12.5. Other Adverse Effects

Other Information : Avoid release to the environment.

SECTION 13: DISPOSAL CONSIDERATIONS

13.1. Waste Treatment Methods

Waste Disposal Recommendations: Dispose of contents/container in accordance with local, regional, national, and international regulations.

Additional Information: Container may remain hazardous when empty. Continue to observe all precautions.

Ecology - Waste Materials: Avoid release to the environment. This material is hazardous to the aquatic environment. Keep out of sewers and waterways.

SECTION 14: TRANSPORT INFORMATION

The shipping description(s) stated herein were prepared in accordance with certain assumptions at the time the SDS was authored, and can vary based on a number of variables that may or may not have been known at the time the SDS was issued.

14.1. In Accordance with DOT Not regulated for transport

14.2. In Accordance with IMDG Not regulated for transport

14.3. In Accordance with IATA Not regulated for transport

SECTION 15: REGULATORY INFORMATION

15.1. US Federal Regulations

Reagent D	
SARA Section 311/312 Hazard Classes	Health hazard - Respiratory or skin sensitization

1,2,3-Propanetriol (56-81-5)
Listed on the United States TSCA (Toxic Substances Control Act) inventory - Status: Active

15.2. US State Regulations

1,2,3-Propanetriol (56-81-5)
U.S. - New Jersey - Right to Know Hazardous Substance List
U.S. - Pennsylvania - RTK (Right to Know) List
U.S. - Massachusetts - Right To Know List

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SECTION 16: OTHER INFORMATION, INCLUDING DATE OF PREPARATION OR LAST REVISION

Date of Preparation or Latest Revision : 11/15/2021

Other Information : This document has been prepared in accordance with the SDS requirements of the OSHA Hazard Communication Standard 29 CFR 1910.1200

This information is based on our current knowledge and is intended to describe the product for the purposes of health, safety and environmental requirements only. It should not therefore be construed as guaranteeing any specific property of the product.

SDS US (GHS HazCom)

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