Doc. 2014A

PSP-36
Total Saxitoxins
Test Kit

Colorimetric Immunoassay
for the detection of
Paralytic Shellfish Poison
in environmental samples

Instructions and User Guide

FOR SCIENTIFIC RESEARCH USE

Manufactured by
Mercury Science Inc.
www.mercuryscience.com
Tel: (866) 861-5836

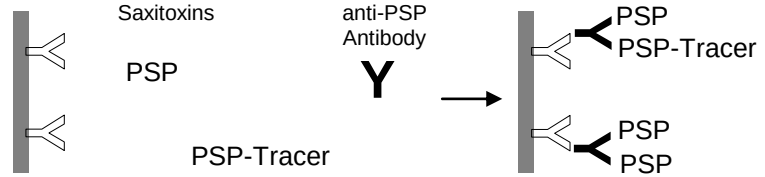
PSP-36
Total Saxitoxins Test Kit

For Scientific Research Use Only.
This product is not to be used for In Vitro or In Vivo Diagnosis.

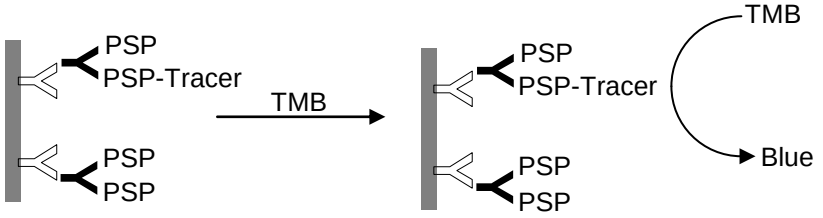
PRINCIPLES OF THE ASSAY
This product contains an antibody (Ab) that binds the toxins responsible for Paralytic Shellfish Poisoning. The assay can be used for for the semi-quantitative detection of total saxitoxins in sample extracts. The signal of samples and a control are compared to determine the amount of saxitoxins present.

The PSP-36 assay is a solid phase colorimetric immunoassay, based on competition between saxitoxin (PSP) and enzyme-labeled saxitoxin (PSP-Tracer) for anti-PSP antibody. Samples containing PSP inhibit the binding of the PSP-Tracer to the antibody molecules. Both the Ab-PSP and Ab-PSP-Tracer complexes are captured on the surface of the microtiter plate wells.

Following a wash step, the addition of an enzyme substrate (TMB) produces a color proportional to the amount of PSP-Tracer in the well. The amount of color measured is inversely proportional to the concentration of toxic saxitoxins in the sample.



Solid phase
anti-mouse IgG



TEST KIT CONTENTS Each PSP-36 test kit contains reagents for testing a maximum of 36 samples in duplicate.
The expiry date of the test kit is stated on the outer label.
Store the kit between 2°C and 8°C.

Reagents Store the reagents between 2°C and 8°C when not in use.

Component	Quantity	
Control Solution	1 vial	2 mL

The control is a phosphate-buffered salt solution with casein.
Contains sodium azide as a preservative.

Sample Dilution Buffer	1 bottle	50 mL

Ready-to-use phosphate buffered (pH 7.8) salt solution with casein.
Contains sodium azide as a preservative.

PSP- Tracer	1 vial	7.5 mL

The tracer is in a MOPS-buffered solution containing bovine protein as a stabilizer and methylisothiazolone, bromonitrodioxane, and Proclin 300 as preservatives.

Anti-PSP Antibody	1 vial	7.5 mL

The antibody is in phosphate-buffered salt solution with casein.
Contains sodium azide as a preservative.

Wash Concentrate*	1 bottle 40 mL

A 25-fold concentration of Tris-HCl buffered (pH 7.8) salt solution with Tween 20. Contains sodium azide as a preservative.

**Prepare for use by mixing entire contents with 960 mL of distilled water and placing in platewasher WASH Bottle.*

Substrate Solution	1 bottle	15 mL

Tetramethylbenzidine (TMB) and H_2O_2

Keep away from direct sunlight.

Stop Solution	1 bottle	15 mL

1 N Hydrochloric Acid

Anti-Mouse IgG Microtitration Strips	1 plate (12 x 8 wells)

WARNINGS AND PRECAUTIONS
For research use only. Handle all samples as potentially hazardous.
Disposal of all waste should be in accordance with local regulations.

SCREENING ASSAY PROCEDURE

Perform each determination in duplicate for the Control and unknowns. All sample extracts should be filtered prior to analysis. All reagents and samples should be brought to room temperature prior to use. Use only the number of strips needed. Keep unused strips stored in their aluminum foil pouch with the included desiccant until needed.

1. Pipet 50 uL of the anti-PSP Antibody solution into each well.

2. Pipet 50 uL of each Control solution or sample into a well using the sequence shown in the table below. **Always use wells A and B on each strip as Controls.** Always perform duplicate analyses of samples. Three samples can be tested per strip. The example below shows the testing of eight samples.

	1	2	3	4	5	6	7	8	9	10	11	12
A	Con-trol	Con-trol	Con-trol									
B	Con-trol	Con-trol	Con-trol									
C	1st Unk	4th Unk	7th Unk									
D	1st Unk	4th Unk	7th Unk									
E	2nd Unk	5th Unk	8th Unk									
F	2nd Unk	5th Unk	8th Unk									
G	3rd Unk	6th Unk										
H	3rd Unk	6th Unk										

3. Shake the wells for 30 minutes.

4. Pipet 50 uL of the PSP-Tracer solution into each well.

5. Shake the wells for 30 minutes.

6. Wash the strips 3 times on the platewasher. Tap the strips upside-down firmly on a paper towel to blot away any excess wash solution that may remain in the wells.

7. Add 100 uL of Substrate Solution to each well. Shake the plate for five minutes.

8. Add 100 uL of Stop Solution to each well. Shake the plate briefly.

9. Measure the absorbance in each well. Note: If Control absorbance is greater than 3.0 AU, remove 50 uL from ALL WELLS and measure absorbance.

10. The data can be analyzed using the Excel worksheet available upon request.

Additional Information

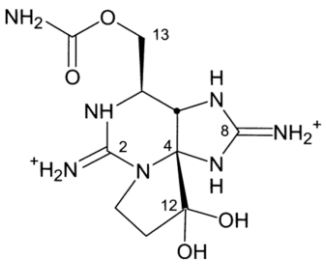
MATERIALS REQUIRED BUT NOT SUPPLIED WITH THE KIT
The PSP-36 test kit is part of a complete system of immunodiagnostic reagents and instrumentation. The system requires the following equipment.

1. Microtiterplate Reader able to measure Absorbance at 450 nm
2. Platewasher
3. Plate Shaker
4. 8 Channel pipet
5. Pipetmen (P10, P200 and P1000)

Other Notes:

- Perform each Control and Sample in duplicate wells.
- All sample extracts should be filtered and diluted prior to analysis.
- All reagents and samples should be brought to room temperature prior to use.
- Use only the number of strips needed.
- Keep unused strips stored in their aluminum foil pouch with the included desiccant until needed.
- If Control absorbance is greater than 3.0 AU, remove 50 uL from ALL WELLS and repeat absorbance measurement.

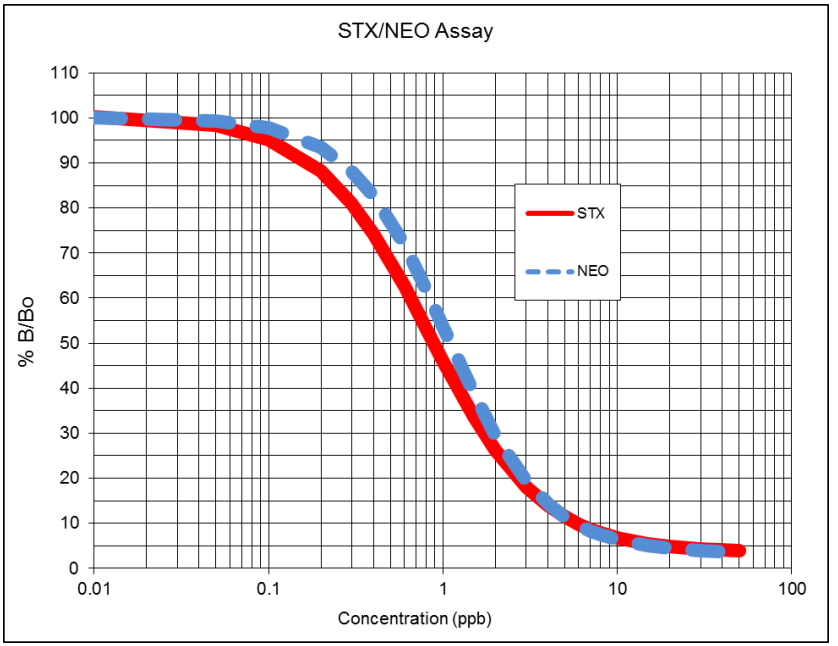
An Excel worksheet has been developed to analyze results and quantitate the amount of Total Saxitoxin in extracts.
Send your request for the "Total Saxitoxin Quantitation Worksheet - PSP-36" to:
info@mercuryscience.com



saxitoxin, STX

PERFORMANCE CHARACTERISTICS

Sensitivity
Saxitoxin and Neosaxitoxin Standard Curves



PERFORMANCE CHARACTERISTICS (Cont.)

Detection Limit

The detection limit is defined as the minimum concentration of Saxitoxin that can be distinguished from a blank standard with 95% confidence. A detection limit of 0.1 ppb Saxitoxin in extraction buffer has been demonstrated with this assay. The quantitative range of the assay is from 0.2 ppb to 6 ppb Saxitoxin.

Cross Reactivity

This assay is specific for the detection of Saxitoxin. The ability of the assay to detect structurally related compounds is shown in the following table.

Analyte	% Reactivity
Saxitoxin	100
Neosaxitoxin	73
GTX3	32
GTX2	25
GTX6(B2)	23
dcSTX	19
GTX4	16
GTX1	15

PROCEDURAL NOTES

Please read all instructions thoroughly before using this kit. Do not mix reagents from kits having different lot numbers. Do not use kits after the expiration date printed on the kit label.

Reagents should be at room temperature when used.

During washing steps, check that each well is completely filled during wash solution additions. After washing is complete, invert the wells and tap them firmly against a paper towel to remove excess liquid.

The platewasher should be rinsed with distilled water at the end of each day of use to prevent clogging of the dispensing and aspirating ports. Prime the platewasher with wash solution before the first wash each day.

Care must be taken during each step to prevent contamination of reagents and equipment. Do not use the same pipet tip in two different reagents.

For Technical Assistance, contact Mercury Science Inc: (866) 861-5836.

Total Saxitoxins Test Kit

Summary Protocol Sheet

Add Antibody	**50 uL**
Add Control and Samples	**50 uL**
Incubate	**Shake for 30 minutes**
Add Tracer	**50 uL**
Incubate	**Shake for 30 minutes**
Wash	**"3 WASHES" program**
Add Substrate	**100 uL, Shake for 5 minutes**
Add Stop	**100uL**
Measure	**Absorbance at 450 nm**

Note: If Control absorbance is greater than 3.0 AU, remove 50 uL from ALL WELLS and repeat absorbance measurement.