STRATEGIC DIAGNOSTICS INC.
EnviroGard® Triazine Test Kit
72100

Intended Use
The EnviroGard Triazine Test Kit is a qualitative "yes/no" or semi-quantitative test for the detection of triazine residues in water or other aqueous solutions at 0.1 and 1.0 ppb. You can use the test kit in the laboratory or on-site without any special training. With the EnviroGard Triazine Test Kit, you can read results visually or perform a more precise analysis with a photometer.

Test Principles
The EnviroGard Triazine Test Kit is based on the use of polyclonal antibodies that bind either triazine or an atrazine-enzyme conjugate. Triazine in the sample competes with the atrazine-enzyme conjugate for a limited number of antibody binding sites. Antibodies that bind triazine are immobilized to the walls of the test tubes.

Since there are the same number of antibody binding sites on every test tube and each test tube receives the same number of atrazine-enzyme conjugate molecules, a sample that contains a low concentration of triazine enables the antibody to bind many atrazine-enzyme conjugate molecules.

Therefore, a low concentration of triazine produces a dark blue sample. Conversely, a high concentration of triazine will allow fewer atrazine-enzyme conjugate molecules to be bound by the antibodies, resulting in a lighter blue sample.

NOTE: Color is inversely proportional to triazine concentration.

Darker color = Lower concentration
Lighter color = Higher concentration

Performance Characteristics
The EnviroGard Triazine Test Kit does not differentiate between atrazine and closely related compounds, but detects their presence to differing degrees. The following chart shows the approximate Lower Limit Of Detection (LLD), for the cross-reacting compounds. Concentration is in parts per billion (ppb).

Compound	LLD
Ametryn	0.063
Atrazine	0.053
Prometon	0.044
Prometryn	0.076
Propazine	0.001
Terbuthylazine	0.010
Simetryn	0.037
Simazine	0.317
Cyanazine	0.994
6-Hydroxy Atrazine	0.120
Trietazine	0.240
Diazinon	*
De-ethyl Atrazine	0.17
De-isopropyl Atrazine	2.31

* Undetectable below 100 ppb

The following compounds were found to be undetectable at 1000 ppb:

2,4-D	Carbaryl	Maneb
Alachlor	Carbofuran	Methamidophos
Aldicarb	Glyphosate	Metolachlor
Azinphos Methyl	Mancozeb	Triclopyr

Precautions
- Store all test components at 4°C to 8°C (39°F to 46°F) when not in use.
- Do not store test components for more than 8 hours at ambient temperatures (18°C to 27°C or 64°F to 81°F).
- Do not freeze test kit components or expose them to temperatures greater than 37°C (99°F).
- Allow all reagents to reach ambient temperature (18°C to 27°C or 64°F to 81°F) before beginning the test (approximately 30-60 minutes).
- Do not use test kit components after the expiration date.
- Do not use reagents or test tubes from one test kit with reagents or test tubes from a different test kit.
- Because of the rapid kinetics of the EnviroGard Triazine Test Kit, do not analyze more than six test tubes at one time.
- Use approved methodologies to confirm any positive results.
- Do not dilute or alter test reagents or samples not called for in the test procedure; this may give inaccurate results.
- Some solutes and particulates found in untreated ground or surface waters may affect the sensitivity level of this test kit.
- Use a calibrator that has a matrix comparable to your sample if you are testing something other than water.

Materials Provided
The EnviroGard Triazine Test Kit contains the following items:

20 antibody-coated test tubes
1 vial of Negative Control
1 vial of Atrazine- Enzyme Conjugate
1 vial of 0.1 ppb Atrazine Calibrator
1 vial of 1.0 ppb Atrazine Calibrator
1 vial of Substrate
1 vial of Chromogen
1 vial of Stop Solution
1 test tube rack

Materials Needed
You will also need several other items:

- Marking pen for test tubes
- Disposable-tip pipette that will measure 160 microliters (µL)
- Stopwatch or wristwatch with second hand
- Tap or distilled water for washing test tubes
- Pipette that will measure 0.5 mL (for photometric interpretation only)
- Calculator (optional)
- Spectrophotometer (optional)

Perform the Test

1. Remove the test tubes from the plastic bag and mark them as follows:

Tube Marking	Tube Contents
"–"	Negative Control
"+"	0.1 ppb or 1.0 ppb Calibrator
"S1"	Sample 1
"S2"	Sample 2
"S3"	Sample 3
"S4"	Sample 4

2. Add 160 µL of Negative Control to the "–" test tube.
3. Add 160 µL of the appropriate calibrator to the "+" test tube.

NOTE: You can use both calibrators to approximate the concentration levels of your samples to a more accurate degree. However, you should not use more than six test tubes in one test. If you use both calibrators, label them accordingly ("0.1 ppb" and "1.0 ppb").

4. Add 160 µL of each sample to the corresponding test tube. Immediately add 4 drops (160 µL) of Atrazine-Enzyme Conjugate to each test tube. Gently swirl the test tubes to mix for 2 to 3 seconds.
5. Leave the test tubes undisturbed for 5 minutes, then shake out their contents.
6. Fill the test tubes to overflowing with tap or distilled water, then decant and vigorously shake out the remaining water.

NOTE: Repeat this wash step three more times, being certain to shake out as much water as possible on each wash.

7. Add 4 drops of Substrate to each test tube. Immediately add 4 drops of Chromogen to each test tube. Gently mix the test tubes for a few seconds.

CAUTION: Do not reverse this order; add the Substrate *before* the Chromogen.

Interpret the Results

You can either interpret the results visually within 2 minutes after adding the substrate and chromogen to each test tube or you can perform a more precise analysis with a photometer.

NOTE: If a blue color does not develop in the negative control test tube within 2 minutes after adding the substrate and chromogen, the test is invalid and you must repeat it.

Visual Interpretation

1. Compare the sample test tube to the negative control test tube against a white background. The test tube rack in the kit is well-suited for this purpose.
2. If the sample test tube contains less color than the negative control test tube, then the original sample contains triazine or a cross-reactant (or combination of the same) at a concentration greater than or equal to those levels indicated in the "LLD" table. (See "Performance Characteristics.")
3. If you are unable to distinguish a difference between the sample and negative control test tubes, pesticides may be present in the sample but at a very low concentration (near the LLD listed). Use a photometer for a more precise analysis.
4. If a sample test tube contains more color than a calibrator, then the original sample contains a lower concentration than that calibrator. Conversely, if a sample test tube contains less color than a calibrator, the sample contains a greater concentration than the calibrator.

Photometric Interpretation

After 2 minutes, add 1 drop of Stop Solution to each test tube and mix well. This turns the solution yellow.

WARNING: Stop Solution is 2.5 N sulfuric acid.

Proceed to the set of instructions appropriate for your type of photometer.

Conventional Spectrophotometers

1. Add 0.5 mL of water to each test tube and gently mix.
2. Adjust the wavelength of your photometer to 450 nanometers (nm) and zero against a water blank.
3. Transfer the reaction liquid to an appropriate cuvette or aspirate directly into the photometer (depending on your photometer type).
4. Measure and record the absorbance (optical density [OD]) of the negative control, each sample, and the calibrator.
5. Compare the OD of each sample to the OD of the negative control. If the OD of the sample is less than 90% of the OD of the negative control, the sample contains a concentration of triazine or related compound greater than or equal to those levels indicated in the LLD table. (See "Performance Characteristics.") If the OD of the sample is greater than 90% of the OD of the negative control, the sample contains less than the LLD limits listed in the same table.
6. Compare the OD of each sample to the OD of the calibrator. If the OD of the sample is less than the OD of the calibrator, the sample contains a concentration greater than the concentration of the

calibrator. Conversely, if the OD of the sample is greater than the OD of the calibrator, the sample contains a concentration that is lower than the concentration found in that calibrator.

Differential Photometer

1. Add 0.5 mL of water to each test tube and gently mix.
2. Place a test tube containing water in the left (reference) well.
3. Place the negative control test tube into the right (sample) well. Record the OD of the sample.
4. Remove the negative control test tube and replace it with the next tube (calibrator or sample) to reactivate the photometer. Record the result. Repeat this procedure to determine the OD for each of the remaining samples.
5. If the OD of the sample is less than 90% of the OD of the negative control, the sample contains a concentration of triazine or related compound greater than or equal to those levels indicated in the LLD table. (See "Performance Characteristics.") If the OD of the sample is greater than 90% of the OD of the negative control, the sample contains less than the LLD limits listed in the same table.
6. Compare the OD of each sample to the OD of the calibrator. If the OD of the sample is less than the OD of the calibrator, the sample contains a concentration greater than the concentration of the calibrator. Conversely, if the OD of the sample is greater than the OD of the calibrator, the sample contains a concentration that is lower than the concentration found in that calibrator.

Example (for illustration only)

	OD	Interpretation
Negative Control	0.994	
0.1 ppb	0.736	
1.0 ppb	0.267	
Sample	0.532	> 0.1 ppb; < 1.0 ppb

Ordering Information

Description	Catalog Number
EnviroGard Triazine Test Kit	72100

Technical Assistance

Strategic Diagnostics Inc.
111 Pencader Drive
Newark, Delaware 19702-3322 USA
800-544-8881
302-456-6789 Phone
302-456-6782 Fax
www.sdix.com techservice@sdix.com

General Limited Warranty

Strategic Diagnostics Inc. warrants the products manufactured by it against defects in materials and workmanship when used in accordance with the applicable instructions for a period not to extend beyond a product's printed expiration date. **SDI MAKES NO OTHER WARRANTY, EXPRESSED OR IMPLIED. THERE IS NO WARRANTY OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE.**

3099450 Rev. 3 FEB 98