

Nebraska Department of Water, Energy, and Environment

Standard Operating Procedure (SOP)

Groundwater Division - GW

SOP Number: GW-130

Title: Field Screening for Triazines in Groundwater

Written Date: July 2025

Purpose: To achieve an accurate field determination of the presence of triazine herbicides in ground water samples for background and public information, and to use as a decision aid in obtaining ground water samples for pesticide analysis.

Equipment/Materials Needed:

- Ground water in a clean unmarked 1-liter plastic container or wide-mouth glass jar as per SOP # GW-060
- Triazine immunoassay field screening kit, including antibody tubes and reagents
- Marking pen for tubes
- Disposable pipettes, polyethylene or equivalent
- Stopwatch or wristwatch with second hand
- Squirt bottle filled with deionized water

Procedures:

NOTE: for more information, consult the manufacturer's instructions supplied with the test kit.

1. Obtain a ground water sample in a clean unmarked 1-liter plastic container or wide-mouth glass jar as per SOP # GW-060.
2. Remove two tubes from the plastic bag in the triazine kit and mark one with a "-" (for negative control) and one with an "S" (for sample).
3. Using a disposable pipette, add four (4) drops of the negative control reagent to the "-" tube.
 - a. (NOTE: this screen requires 160 µl of negative control and sample water, or the same as 4 drops from the reagent bottles. If there is any doubt that the pipette you are using is dispensing approximately the same amount as the reagent bottles, use a graduated cylinder or other device to determine how many drops from the pipette you are using will give an approximately equal amount).
4. Dispose of the pipette in an ordinary waste container.
5. Using a disposable pipette, add four (4) drops of sample water to the "S" tube.
6. Dispose of the pipette in an ordinary waste container.
7. Immediately add four (4) drops of enzyme conjugate to each tube.
8. Swirl the tubes gently to mix for 2-3 seconds.
9. Stand tubes in the plastic tube rack in an appropriate place and leave them undisturbed for five (5) minutes, marking the time with a wristwatch or stopwatch.
10. After the five minutes have elapsed, shake out the contents of the tubes.
11. Fill the tubes to overflowing with deionized water, then dump out and vigorously shake out the remaining water. Repeat this wash step three (3) more times, being certain to shake out as much water as possible on each wash.
12. Add four (4) drops of substrate reagent to each tube.
13. Follow IMMEDIATELY with four (4) drops of chromogen reagent to each tube. Be sure not to reverse the order of these reagents.
14. Swirl the tubes to mix gently for a few seconds.
15. Read the results visually within 2 minutes. The negative control ("-") tube should develop a blue color; the sample ("S") tube may or may not develop any color. REMEMBER: color is inversely

proportional to concentration; that is, darker color = lower concentration, and lighter color = higher concentration.

16. If there is ANY color difference between the “-” and “S” tubes (i.e., if the “S” tube sample is ANY lighter than the “-” tube sample), this indicates the presence of triazine herbicides. If this is the case, proceed with taking a ground water sample for pesticide analysis as per SOP # GW-060.
17. Vigorously shake out the tubes and dispose of them in an ordinary waste container.
18. Reassemble the test kit and other materials, and secure them for transport to the next sample site.