

Read instructions carefully before starting test

Reveal[®] 2.0 **for ASP**

For use with approved NEOGEN reader
Store at 18–30°C (64–86°F) • Do not freeze

THE TOXINS

Toxins that cause amnesic shellfish poisoning (ASP) are produced by toxigenic diatoms of the genus *Pseudo-nitzschia*. ASP toxins primarily include domoic acid (DA). In addition to contamination of seafood, these marine biotoxins can result in human and marine wildlife mortality. The clinical toxicological effects attributed to DA can include: permanent loss of short-term memory, nausea, vomiting, headache, disorientation and loss of balance. Action limits for DA were established soon after the 1987 domoic acid/mussel crisis in Canada, which included more than 100 cases of human illnesses and also several mortalities as a direct result of contaminated shellfish consumption. Most countries have currently established a maximum permitted level of 20 mg DA per kg whole shellfish (20 ppm).

INTENDED USE/USER

Reveal[®] 2.0 for ASP is intended for the qualitative screening of shellfish for DA, which produces a positive result with samples containing 20 ppm or above. The test kit is designed for use by quality control personnel and other personnel familiar with handling shellfish possibly contaminated by DA toxins.

ASSAY PRINCIPLES

Reveal 2.0 for ASP is a single-step lateral flow device based on a competitive immunoassay format. Shellfish extract is wicked through a reagent zone containing antibodies specific for DA which have been conjugated to colored particles. If DA is present in the sample, the toxin will be captured by the particle-antibody complex. The complex then is wicked onto a membrane, which contains a stationary capture zone of a toxin-protein conjugate. This zone captures any uncomplexed DA particle-antibody. Therefore, as the concentration of DA in the sample increases, the test line intensity decreases. The membrane also contains a stationary control zone which will always form regardless of the level of DA.

STORAGE REQUIREMENTS

Store kit components at room temperature (18–30°C, 64–86°F) to ensure full shelf life. Test strips should remain capped in their original sample tubes until used to ensure optimal performance.

MATERIALS PROVIDED

Reveal 2.0 for ASP (Neogen item 9560)

1. 24 Reveal 2.0 for ASP lateral flow test strips
2. 24 microwells
3. 24 vials of ASP buffer
4. 25 extraction bags
5. 48 disposable 100 µL pipettors

MATERIALS RECOMMENDED BUT NOT PROVIDED

1. Marine Biotoxins Starter Kit (NEOGEN item 9563)
 - a. Microwell holder
 - b. 1 roller
 - c. 1 bag clip (white clip and green straw)
2. Distilled water
3. Sample collection cups with lids (NEOGEN items 9428, 9428B)
4. Blender (NEOGEN items 9493, 9477 or 9495)
5. Scale capable of weighing 0.5–400 g ± 0.1 g (NEOGEN item 9427)
6. Timer (NEOGEN item 9452)
7. Graduated cylinder, 50 mL (Neogen item 9367) or bottle-top dispenser (NEOGEN item 9448)
8. AccuScan Pro reader (NEOGEN item 9565) or Raptor SOLO Integrated Analysis Platform (NEOGEN item 9696)
9. Raptor Cartridges (Neogen item 9681)
10. Raptor Exact Volume Transfer Pipettes (NEOGEN item 9682)

PRECAUTIONS

1. The test strips must remain inside the stay dry tube before use.
2. Store test kit at room temperature (18–30°C, 64–86°F) when not in use. Do not freeze.
3. Do not use kit contents beyond expiration date.
4. Treat all liquids, including sample extract, and used components as if contaminated with toxin. Gloves and other protective apparel should be worn at all times.
5. To avoid cross-contamination, use clean pipettors, extraction bags and fresh extraction solutions for each sample.

ACCUSCAN PRO READER SET UP

1. Enter the lot-specific QR code by selecting the QR code icon on the reader. Place the QR code into the cartridge and insert the cartridge into the reader.
NOTE: For instructions on manually entering sample IDs, see the AccuScan Pro user manual.
2. Return to the home screen and select the test strip icon. Touch the **Marine Biotoxins** category, then select the **ASP** test type.

SAMPLE PREPARATION AND PRELIMINARY EXTRACTION

The sample to be tested should be collected according to accepted sampling techniques.

1. Obtain a representative sample. Shell the samples.
2. Thoroughly rinse the samples with distilled or deionized water, and allow any excess water to drain.
3. Homogenize (e.g., blend, puree) the shellfish in a high-speed blender.
NOTE: A good homogenate is essential in order to obtain an accurate result.
4. Weigh 1 g (\pm 0.05 g) of homogenized sample in a sample cup.
5. Pour 30 mL (\pm 0.5 mL) of distilled water into sample cup containing the sample and secure the lid.
6. Shake the sample cup vigorously by hand for **30 seconds**, until all shellfish tissue is in solution (a cloudy appearance or bubbles may form, which does not affect the running of the test).
7. Number both sides of an extraction bag using a marker, so that there is a side labeled “1” and the other side labeled “2.” Pour solution/sample mixture into the side labeled “1.”
NOTE: The extraction bag contains a mesh filter which allows for partial filtration of the sample. All samples/solutions should only ever be added to the side labeled “1.”
8. To seal the bag, position and hold the green straw approximately 2–3 inches down from the top of the bag, fold the upper edge of the bag so that it covers the green straw and firmly clip on the white bag clip. This prevents leakage of the sample.
9. Press the roller firmly on the sample extraction bag, pushing the roller back and forth for **30 seconds** to aid in obtaining a homogeneous sample extract.
10. Slide out the green straw and remove the white bag clip.
11. Pour all the bag contents from side “2” back into the original sample cup (there may be small pieces of shellfish remaining on side “1” of the bag). Discard the used extraction bag.
12. Cap and shake the sample cup vigorously by hand for **30 seconds** (a cloudy appearance or bubbles may form, which does not affect the running of the test).
13. Remove 100 μ L of the sample extract using a disposable pipettor* provided (or alternatively by use of a standard pipettor), and add into an ASP buffer vial.
*To use the disposable pipettors, firmly press the top bulb of the pipettor, insert the tip into the solution, slowly release the top bulb to draw up the sample extract. Excess volume (e.g., more than 100 μ L) will overflow into the lower bulb, ensuring 100 μ L is ready to dispense. Press the top bulb firmly and release slowly to dispense. Discard the used pipettor.

TEST PROCEDURE

1. Remove the appropriate number of microwells and place into the microwell holder.
2. Shake the ASP buffer vial (containing diluted sample) vigorously by hand for **30 seconds**.
3. Immediately transfer 100 µL of diluted sample into each microwell using a new disposable pipettor.
4. Remove the required number of test strips from the lateral flow device container and immediately close the container tightly.
5. Place the ASP test strips with the sample end down (Neogen logo on top) into the microwells.
6. Allow the strip to develop in the microwell for **10 minutes**.
7. Immediately remove the test strip and read using the Accuscan Pro reader (as described below).

READING TEST RESULTS

Test strips should be read within **1 minute** of completion of the 10 minute incubation. Refer to **AccuScan Pro Reader Set Up** for test selection and set up information.

1. Fully insert the Reveal 2.0 for ASP test strip into the black cartridge adapter with the sample end first and results facing out.



2. Insert the cartridge with test strip side up in the AccuScan Pro. The reader will automatically begin analyzing the cartridge. **CAUTION:** Removing cartridge prior to completion can result in invalid readings.
3. The AccuScan Pro reader will analyze the test strip and results will be displayed and stored in the reader.



NOTES

1. Ensure device is fully inserted into cartridge.
2. Readings should be made between **10–11 minutes**. Readings after 11 minutes may be inaccurate due to over-development of the device.
3. The strips must be read using a Neogen-approved reader.

TEST PROCEDURE – RAPTOR SOLO INTEGRATED ANALYSIS PLATFORM

1. Fully insert a Reveal for 2.0 for ASP test strip into a Raptor cartridge. Up to 3 strips can be inserted into the Raptor cartridge at one time to obtain duplicate or triplicate results.
2. Insert the Raptor cartridge containing the test strip(s) into the port within the Raptor SOLO Integrated Analysis Platform reader.
3. The bar code on the test strip(s) will be read — the Raptor SOLO reader identifies the type of test strip and the lot number.
4. If the lot number is not found in the system, the bar code reader on the front of the Raptor will turn on automatically.
5. Scan the QR code found on the side of the tube containing the test strips. The information will be stored on the system.

6. Enter Sample ID if desired.
7. Shake the ASP buffer vial (containing diluted sample) vigorously by hand for **30 seconds**.
8. Immediately transfer 400 μ L of diluted sample into the Raptor cartridge using a new disposable pipettor.
9. The Raptor SOLO reader will start automatically.
10. Results will be displayed on the Raptor screen after the **10 minute** testing is complete.

PERFORMANCE CHARACTERISTICS

All Neogen-approved readers will report **Positive** with a result of 20 ppm or greater. Any result of less than 20 ppm will be reported as **Negative**.

VALIDATED MATRICES

Mussels, scallops, oysters, clams and cockles.

NOTE: Neogen continues to validate new commodities. Please contact a representative for the latest validated commodity list.

CUSTOMER SERVICE

Neogen Customer Assistance and Technical Services can be reached by using the contact information on the back of this booklet. Training on this product, and all Neogen test kits, is available.

SDS INFORMATION AVAILABLE

Safety data sheets (SDS) are available for this test kit, and all of NEOGEN's test kits, on NEOGEN's website at neogen.com, or by calling NEOGEN at 800.234.5333 or 517.372.9200.

TERMS AND CONDITIONS

For NEOGEN'S full terms and conditions, please visit neogen.com/terms-and-conditions/

WARRANTY

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TESTING KITS AVAILABLE FROM NEOGEN

Natural toxins

- Aflatoxin, DON, ergot alkaloids, ochratoxin, zearalenone, T-2/HT-2 toxins, fumonisin, histamine

Foodborne bacteria

- *E. coli* O157:H7, *Salmonella*, *Listeria*, *Listeria monocytogenes*, *Campylobacter*, *Staphylococcus aureus*, *Salmonella enteritidis*

Sanitation

- ATP, yeast and mold, total plate count, generic *E. coli* and total coliforms, protein residues

Food allergens

- Almonds, coconut, crustaceans, eggs, gliadin, hazelnut, milk, mustard, peanuts, sesame, soy, walnuts, multi-treenut

Genetic modification

- CP4 (Roundup Ready®)

Ruminant by-products

- Meat and bone meal, feed

Species identification

- Raw and cooked meat samples



North America
NEOGEN Headquarters
800.234.5333 (USA/Canada)
foodsafety@neogen.com
NEOGEN.com

Europe, Middle East and Africa
NEOGEN Europe
+ 44 (0) 1292 525 600
info_uk@neogeneurope.com
NEOGEN.com

Mexico
NEOGEN Latinoamerica
+52 (55) 5254-8235
informacion@neogenlac.com
NEOGEN.com

Brazil
NEOGEN do Brasil
+55 19 3935.3727
info@neogendobrasil.com.br
NEOGEN.com

China
NEOGEN Bio-Scientific Technology
+86 21 6271 7013
info@neogenchina.com.cn
www.neogenchina.com.cn

India
NEOGEN Food and Animal Security
+91 484 2306598, 2301582
info@neogenindia.com
www.neogenindia.com