United States Department of Agriculture
Agriculture Marketing Service
Federal Grain Inspection Service

# Test Kit Instruction

June 22, 2021

## NEOGEN
## REVEAL Q+ FOR DON
## USING RAPTOR INTEGRATED ANALYSIS PLATFORM

### FORWARD

The instructions presented in this document cover only the procedure for performing the analytical test for official inspections. For questions regarding this procedure, contact Dr. Ajit Ghosh of the Technology and Science Division by phone at 816-702-3817 or email at Ajit.K.Ghosh@usda.gov.

Refer to the Mycotoxin Handbook for information on use of this test kit in the official inspections including sampling, general sample preparation, reporting and certification of test results, laboratory safety, and hazardous waste management. For questions regarding these policies and/or instructions, contact Patrick McCluskey (816-702-3923 or email at Patrick.J.McCluskey@usda.gov.

# Contents

## 1. GENERAL INFORMATION

The Reveal Q+ for DON test method provided by the Neogen is a single- step lateral flow immunochromatographic assay based on a competitive immunoassay format. The test provides quantitative analysis for the presence of DON, using water as an extraction solvent along with a DON-antibody particle complex coated test strip and the Neogen Raptor Integrated Analysis Platform.

| **Approved Test Kit Information** | |
|---|---|
| **Test Kit Vendor:** | Neogen 800-234-5333 |
| **Test Kit Name:** | Reveal Q+ for DON using the Raptor Integrated Analysis Platform |
| **Product Number:** | 8385 |
| **Effective Date of Instructions:** | 6/22/2021 |
| **Conformance Range:** | 0.50 – 30 ppm |
| **Number of Analyses to Cover Conformance Range:** | 2 |
| **Type of Service:** | Quantitative |
| **Approved Commodities:** | Corn (field/dent corn, corn meal, cracked corn, corn flour, corn grits/polenta, corn screenings), wheat (whole grain wheat flour, wheat middlings, wheat red dog, wheat flour 2nd clear, and wheat screenings). |
| **Extraction method:** | Shake 50 grams sample with 250 milliliter (mL) of deionized or distilled water on a mechanical shaker at 250 rpm (or by hand with similar shaking action) for 3 minutes. |
| **Test Format:** | Lateral Flow Strip |
| **Detection Method:** | Raptor Integrated Analysis Platform, Model #9680 |

## 2. PREPARATION OF TESTING MATERIALS

a. **Raptor Integrated Analysis Platform Set-Up:**

The system provides an easy method to objectively read, store, and analyze results from NEOGEN's line of lateral flow strips.

N**ote**: Please keep and store all packaging materials included in the kit for future storage.

(1) Fully insert a Reveal Q+ DON test strip into a Raptor cartridge.

(2) Insert the Raptor cartridge containing the test strip into any of the three ports within the Raptor.

(3) The bar code on the test strip will be read. The system identifies the type of test strip and the lot number. If the lot number is not found in the system, the bar code reader in the front of the Raptor will turn on automatically.

(4) Scan the QR code found on the tube containing the test strips. The information will be stored on the Raptor.

(5) Enter Sample ID if desired.

## 3. SAMPLE PREPARATION AND EXTRACTION PROCEDURES

The sample to be tested should be collected and prepared according to accepted sampling techniques (see **Mycotoxin Handbook for details**).

a. **Extraction Procedure:**

(1) Weigh 50 g (± 0.2 g) ground samples into a Whirl-Pak bag.

(2) Using a 250 mL graduated cylinder, add 250 mL of distilled or deionized water and close the bag securely to prevent spillage.

(3) Shake vigorously on a mechanical shaker at 250 rpm (or by hand with similar shaking action) for 3 minutes. Allow the sample to settle for a minimum of 3 minutes.

(4) Filter at least 3 mL of the extract using a syringe filter (Neogen item #9420) into a clean collection tube labeled with the sample identification. The filtrate should be free of any particulates. If the filtrate is cloudy, perform a second filtration.

(5) Dilute the filtered extract 3:2 by adding 900 microliters (µL) of the filtered extract (measured using a 1000 µL adjustable pipettor) to 600 µL of distilled or deionized water (measured using a 1000 µL adjustable pipettor) in a new test tube. Vortex for 10 seconds. This is the **diluted filtered extract** and is ready for testing, which can be used for next 4 hours if capped. Vortex for a few seconds before the analysis.

(6) Proceed to T**EST PROCEDURES** section below.

## 4. TEST PROCEDURES

**NOTE:** For all unknown samples, analysis procedure 4a should be followed first. If the result is above 5 ppm, proceed to analysis procedure 4b.

a. **Analysis Procedure (0.50 – 5.0 ppm Quantitation Range)**

(1) Place the appropriate number of red sample dilution cups for each test sample in the sample cup rack. Label cups if necessary.

(2) Using a 1000 µL pipettor, add 1000 µL of sample diluent to each red sample dilution cup.

(3) Using a 100 µL pipettor, add 100 µL of the **diluted filtered extract** from section 3a (5) above into each red dilution cup containing 1000 µL sample diluent. Mix by swirling with the pipette tip first and then by pipetting up and down at least 5 times.

(4) Fully insert a Reveal Q+ DON test strip into a Raptor cartridge.

(5) Insert the Raptor cartridge containing the test strip into any of the three ports within the Raptor Integrated Analysis Platform.

a. The bar code on the test strip will be read. The system identifies the type of test strip and the lot number. If the lot number is not found in the system, the bar code reader in the front of the Raptor with turn on automatically.

b. Scan the QR code found on the tube containing the test strips. The information will be stored on the Raptor.

(6) Using an adjustable 100 – 1000 µL pipettor or a fixed 400 µL pipettor, first mix by pipetting up and down a couple of times and then transfer 400 µL to the Raptor cartridge. Press Next. Raptor system will start automatically and the result will be displayed upon the completion of 4 minute incubation.

(7) Additional samples can be analyzed in the other ports while the

first sample is processing.

(8) Results following this protocol are valid in the range of 0.5 – 5 ppm. If the result is more than 5 ppm, run the test using extraction procedure for 5 –30 ppm quantitation range (**Dilution A Protocol**) below.

b. **Dilution A Protocol (5.0 – 30 ppm Quantitation Range)**

(1) Dilute the **diluted filtered extract** (from 3a(5) above) fifteen-fold with distilled or deionized water as below:

(2) Using a 1000 µL pipettor, add 1400 µL (measure first 1000 µL and then 400 µL) of distilled or deionized water to a test tube.

(3) Using a 100 µL pipettor, add 100 µL of **diluted filtered extract** to the 1400 µL of distilled or deionized water. This is **Diluted Extract A** and is good for 4 hours if capped. Vortex for a few seconds prior to the analysis.

(4) Follow the same test procedure as described in "4**a. Analysis Procedure (0.5 – 5.0 ppm Quantitation Range)**" except instead of 100 µL of the **diluted filtered extract** use 100 µL of the **Diluted Extract A**.

(5) Multiply the results by 15.

A result (using 5.0 – 30 quantitation range) less than 3.5 ppm is indicative of a problem, and troubleshooting is needed. Verify the procedure is being followed properly. Perform the analysis using 0.5 – 5 ppm quantitation range and only perform the analysis using 5 – 30 ppm quantitation range again if the value is greater than 5 ppm.

## 5. REPORTING AND CERTIFYING TEST RESULTS

Refer to the Mycotoxin Handbook for reporting and certification of test results. For questions regarding these instructions, contact Patrick McCluskey (816-702-3923 or Patrick.J.McCluskey@usda.gov).

## 6. STORAGE CONDITIONS AND PRECAUTIONS

a. **Storage Conditions:**

Store test kit at room temperature (18-30°C, 64-86°F) when not in use, do not freeze. Test strips should remain capped in their original tubes until used to ensure optimal performance.

b. **Precautions:**

(1) Do not use test kit components beyond the expiration date.

(2) Ensure the device lot number and the curve details match the lot ID number selected on the reader. Failure to update the lot-specific QR code within the reader will cause inaccurate results.

(3) Treat all used liquids, including sample extract, and lab ware as if contaminated with DON. Gloves and other protective apparel should be worn at all times.

(4) To avoid cross-contamination, use new pipette tip for each measurement.

## 7. EQUIPMENT AND SUPPLIES

a. **Materials provided in test kits:**

(1) 25 Reveal Q+ for DON test strips

(2) 25 clear sample cups and 25 red sample dilution cups

(3) Two bottles of sample diluent

b. **Materials required but not provided:**

(1) Timer (NEOGEN item #9426)

(2) Mechanical Shaker (VWR Catalog #89032-096)

(3) 100 µL pipettor (NEOGEN item #9272- fixed, #9278-Basic fixed)

(4) 1000 µL pipettor (NEOGEN item #9337-fixed)

(5) 400 µL pipettor (NEOGEN item #9693- fixed, #9693MLA- MLA fixed)

(6) 100–1000 µL Pipettor, (NEOGEN item #9290)

(7) 100 µL pipette tips (NEOGEN item #9407-tip rack (96 tips), #9410-bag of 1000 tips, #9417-10 reload decks)

(8) 1000 µL pipette tips (NEOGEN item #9464- bag of 1000 tips, #9487- tip rack (96 tips), #9292- tip rack (5 racks of 192 tips), #9293- bag of 1000 tips)

(9) Whirl Pak Stomacher type sampling bag (NEOGEN item #9736)

(10) Reveal sample rack. (NEOGEN item #9475)

(11) Filter syringe (NEOGEN item #9420)

(12) Sample collection tubes (NEOGEN item #9421)

(13) Raptor Integrated Analysis Platform (NEOGEN item #9680)

(14) Raptor Cartridges (NEOGEN item #9681)

(15) Disposable polyethylene transfer pipettes (Fisher Scientific Cat No. 13-711-9AM)

(16) Dispensing pump (NEOGEN item #9448) or graduated cylinder (NEOGEN item #9368)

(17) Agri-Grind grinder or equivalent (NEOGEN item #9427, see Mycotoxin Handbook for details)

(18) Scale capable of weighing 50 grams (NEOGEN item #9427, see Mycotoxin Handbook for details)

(19) Bottle, 1 Liter (NEOGEN item #9472)

(20) Vortex (NEOGEN item #9494)

## 8. REVISION HISTORY

Effective: 6/22/2021