

Program Notice

FGIS-PN-12-06 08/13/12

REVEAL Q+ FOR DON TEST METHOD

1. PURPOSE

The purpose of this program notice is to provide instructions for the Reveal Q+ DON (vomitoxin) test method, product number 8385, recently approved by the Technology and Science Division for official use.

2. BACKGROUND

The Grain Inspection, Packers, and Stockyards Administration's (GIPSA), Field Management Division (FMD), Policies, Procedures, and Market Analysis Branch (PPMAB), and the Technology and Science Division (TSD) evaluate new test methods, and improvements to previously approved methods in order to provide the market with performance-verified rapid mycotoxin test kits.

In order to offer newly approved/revised test methods in a timely manner, this program notice is issued prior to the release of the revised DON Handbook. The following test procedures are approved for official use by field offices and official service providers.

3. TEST PROCEDURES

a. General Information.

The Reveal Q+ for DON (vomitoxin) test method provided by the Neogen Corporation is a single-step lateral flow immunochromatographic assay based on a competitive immunoassay format. The test provides quantitative analysis for the presence of vomitoxin using a vomitoxin-antibody particle complex coated test strip and the AccuScan III reader with software version 4.22 or newer. The inspector is able to determine the presence of vomitoxin in a sample by following TSD approved test procedures and Neogen's assay principles.

The test kit is limited to providing quantitative DON measurements between 0.5 to 5 ppm. **Test results above the 5 ppm maximum conformance limit must be reported as "exceeding 5 ppm". To report results above 5 ppm, please refer to supplemental analysis procedure section.**

Obtain samples according to the instructions in the Grain Inspection Handbook, Book I “Grain Sampling”, and refer to the DON Handbook Chapter 3 “Sample Preparation”, for minimum sample requirement and sample preparation procedures.”

The extraction solution and other materials used with this test method do not necessitate the use of a separate FGIS-approved laboratory space. Federal Grain Inspection Service (FGIS) employees must comply with all applicable safety and sanitation requirements as listed in the DON Handbook, Chapter 2, “Laboratory Safety”, to ensure a safe and efficient work environment.

Approved Test Kit Information					
Test Kit Vendor:	Neogen Corporation 1-800-234-5333				
Test Kit Name:	Reveal Q+ for DON Test Method				
Product Number	Conformance Limits		Type of Service	Extraction Solution	Supplemental Analysis
8385	Min	Max	Quantitative	Distilled or Deionized water	Yes
QAC Number	0.5 ppm	5 ppm			
VOMT					
Grain/Commodities Approved for					
wheat, corn, barley, malted barley, corn gluten meal, oats, rough rice, and distillers dried grains with solubles.					

- b. Preparation of Extraction Solution.
 - (1) Obtain distilled or deionized water for extraction.
 - (2) Clearly label and store at room temperature in a tightly sealed container.
 - (3) Dispose in accordance with all applicable local, state, and federal disposal regulations.

c. Reveal AccuScan III Reader Set Up.

The AccuScan III reader must have software version 4.22 or newer to analyze the Reveal Q+ tests. Contact Neogen for more information.

- (1) Launch the AccuScan III program on the reader.
- (2) Select the Mycotoxin Q+ category and Q+ DON test.

- (3) Enter a sample ID (optional).
- (4) Enter the test kits lot number and A, B, C, and D values from the test kit box label or select from the history for previously entered lot number details.

Note: Refer to the AccuScan III manual for more detailed instructions. The AccuScan III reader must match the lot number of test strips being analyzed.

****Technicians must update or verify reader information (lot number of test strips in use) before official testing.****

d. Extraction Procedures for: wheat, corn, barley, malted barley, corn gluten meal, oats, rough rice, and distillers dried grains with soluble.

- (1) Transfer 50 grams of ground sample into an extraction mixing jar.
- (2) Add 250 ml of distilled or deionized water.
- (3) Cover the extraction jar shake vigorously for 3 minutes.
- (4) Allow the sample to settle. Filter 5 ml of the extract using a filter syringe and collect the filtrate in a clean sample collection tube labeled with the sample identification.
- (5) After collecting the filtrate (filtered extract), dispose of the filter and ground material according to waste disposal guidelines.

Note: When testing Distillers Dried Grains with Solubles the filtered extract pH level must be verified and adjusted before analysis. If pH is below 6.0 the sample pH needs to be adjusted.

To adjust pH:

- Using a disposable polyethylene transfer pipet, add one drop of 1N NaOH (sodium hydroxide) to the sample extract, vortex or swirl to mix and check the pH with a pH test strip.
- If pH is still under 6.0, add another drop of 1N NaOH, mix, and check pH again. Continue this process until the pH falls between 6.0 - 8.0.

- (6) Dilute the filtered sample extract 1:2 (1 + 1) ratio with distilled or deionized water. For example, add 1 ml of extract to 1 ml of distilled or deionized water.
- (7) The sample extract is now ready for testing.

e. Test Procedures.

(1) Sample Analysis.

- (a) Place the appropriate number of red sample dilution cups and clear sample cups for each test sample in the sample cup rack. Label cups if necessary.
- (b) Using a single-channel pipettor with a new pipette tip, add 1000 microliters (μ l) of sample diluent to each red sample dilution cup.
- (c) Using a new pipette tip, add 100 μ l of sample extract into each red dilution cup with sample diluent. Mix by pipetting up and down 5 times, discard tip.
- (d) Transfer 100 μ l of diluted sample extract into a new clear sample cup.
- (e) Place a new Reveal Q+ for DON test strip with the sample end down into the sample cup and set timer for 3 minutes.
- (f) At the end of the 3 minute incubation/development period, remove the test strip from the sample cup. Proceed directly to reading test results.

(2) Reading Test Results with the AccuScan III Reader.

Test strips should be read within 1 minute after completion of the 3 minute incubation period. Refer to Reveal AccuScan III Reader for set-up and selection information.

- (a) Fully insert the Reveal Q+ test strip into the black cartridge adapter with the sample end first and results facing out.



- (b) Insert the cartridge with test strip upside-down into the reader (the test lines will face downward into the reader).



- (c) The reader's green light will glow when a cartridge is inserted, and will automatically begin analyzing the cartridge.
- (d) The AccuScan III reader will analyze the test strip, and results will be displayed, and stored in the reader.

Notes and Cautions:

- Ensure device is fully inserted into cartridge.
- Removing the cartridge prior to completion can result in invalid readings.
- Reading should be made between 3 and 4 minutes. Reading results after 4 minutes may be inaccurate due to over development of the device.
- The strips must be read using Neogen's Reveal AccuScan III reader.

- (3) Supplemental Analysis Procedures.

- (3) Test results between 0.6 ppm and 5.4 ppm are certified to the nearest whole ppm.
- (4) For test results over 5.4 ppm please follow the supplemental analysis procedure listed above.

Refer to the DON handbook Chapter 4 “Certification”, for more detailed certification procedures.

g. Cleaning Labware.

Clean any reusable labware (e.g., glass collection jars) in a soapy water solution, rinse with clean water, and dry before reusing.

h. Waste Disposal.

After the test has been completed, the remaining sample extracts and sample solutions may be poured down the drain. Discard solid material in the trash can for routine disposal.

i. Equipment and Supplies.

(1) Materials Supplied in Test Kits.

- (a) 25 Reveal Q+ DON test strips.
- (b) 25 red sample dilution cups.
- (c) 25 clear sample cups.
- (d) 2 bottles of sample diluent.

(2) Materials, Supplies and Equipment Recommend but not Provided.

- (a) Timer. (Neogen item #9426)
- (b) 100 µl pipettor. (Neogen item #9860, 9410)
- (c) 500 µl pipettor. (Neogen item #9336, 9464)
- (d) Sample collection cups with lids. (Neogen item #9428)
- (e) Reveal sample rack. (Neogen item #9475)
- (f) Sample Collection tubes with caps. (Neogen item #9421, #9421B)

- (g) Reveal AccuScan III Reader. (Neogen item #9590)
 - (h) Distilled or Deionized Water.
 - (i) Dispensing pump or graduated cylinder. (Neogen item #9448, #9447)
 - (j) Filter Syringe. (Neogen item #9420)
 - (k) Pipette tips, 200-1000 μ l. (Neogen item #9464, #9487)
 - (l) pH test strips. (Neogen item #9478)
 - (m) Disposable polyethylene transfer pipettes.
 - (n) Sample grinder.
 - (o) Scale capable of weighing 5 – 50 grams.
- j. Storage Conditions and Precautions.
- (1) Storage Conditions.
 - (a) Store kit components at room temperature (18 - 30°C, 64 - 86°F) when not in use to ensure full shelf life.
 - (b) Test strips should remain capped in their original tubes until used to ensure optimal performance.
 - (c) Do not freeze test kit components.
 - (2) Precautions.
 - (a) Do not use test kit components beyond expiration date.
 - (b) Test strip development times, other than those specified in Test Procedures, may give inaccurate results.
 - (c) Treat all used liquids, including sample extract, and labware as if contaminated with DON, gloves and other protective apparel should be worn at all times.
 - (d) To avoid cross-contamination, use clean glassware for each sample, and thoroughly wash all glassware between samples.

- (e) Do not use any filter paper for filtering sample extracts. The use of a filter syringe is the only TSD approved filtering procedure for this test kit.

4. FILING

Retain a copy of this program notice with the DON Handbook until the handbook is revised to include the test procedures stated herein.

5. QUESTIONS

Direct any questions concerning this program notice to Carl Jackson, PPMAB, at (202) 720-8286, email carl.jackson@usda.gov or Patrick McCluskey, PPMAB, at (816) 659-8403, email patrick.j.mccluskey@usda.gov.

/s/ Robert Lijewski

Robert Lijewski, Director
Field Management Division