

CALCULATE RESULTS

1. Semi-quantitative results can be derived by simple comparison of the sample absorbances to the absorbance of the calibrator wells: Sample containing less color than a calibrator well will have a concentration of DON greater than the concentration of the calibrator. Samples containing more color than a calibrator well will have a concentration less than the concentration of the calibrator.
2. Quantitative interpretation requires graphing the absorbances of the calibrators (Y axis) versus the log of the calibrator concentration (X axis) on semi-log graph paper. A straight line is drawn through the calibrator points and the sample absorbances are located on the line. The corresponding point on the Y-axis is the concentration of the sample. Samples with absorbances greater than the lowest calibrator or less than the highest calibrator must be reported as < 0.2 ppm or >2.5 ppm, respectively.

SAMPLE CALCULATIONS

Well Contents	OD	Average OD \pm SD*	%RSD	%Bo**	DON Conc. (ppm)
Negative Control	2.714 2.72	2.717 \pm 0.004	0.16	100	N/A
0.2 ppm Calibrator	1.876 1.89	1.883 \pm 0.010	0.53	69.3	N/A
0.5 ppm Calibrator	1.198 1.194	1.196 \pm 0.003	0.24	44	N/A
1 ppm Calibrator	0.799 0.789	0.794 \pm 0.007	0.89	29.2	N/A
2.5 ppm calibrator	0.348 0.353	0.351 \pm 0.004	1.01	12.9	N/A
Sample	0.487 0.497	0.492 \pm 0.007	1.44	18.1	1.727

Actual values may vary; this data is for example purposes only.

* standard deviation

** %Bo equals average sample absorbance divided by average negative control absorbance times 100%.

TECHNICAL ASSISTANCE

For questions regarding this kit or for additional information about Beacon products, call (207) 571-4302.

SAFETY

To receive complete safety information on this product, contact Beacon Analytical Systems, Inc. and request Material Safety Data Sheets. Stop Solution is 1N hydrochloric acid. Handle with care.

General Limited Warranty

Beacon Analytical Systems, Inc. ("Beacon") 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. BEACON MAKES NO OTHER WARRANTY, EXPRESSED OR IMPLIED. THERE IS NO WARRANTY OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE. The warranty provided herein and the data, specifications and descriptions of Beacon products appearing in published catalogues and product literature may not be altered except by express written agreement signed by an officer of Beacon. Representations, oral or written, which are inconsistent with this warranty or such publications are not authorized and, if given, should not be relied upon.

In the event of a breach of the foregoing warranty, Beacon's sole obligation shall be to repair or replace, at its option, any product or part thereof that proves defective in materials or workmanship within the warranty period, provided the customer notifies Beacon promptly of any such defect. The exclusive remedy provided herein shall not be deemed to have failed of its essential purpose so long as Beacon is willing and able to repair or replace any nonconforming Beacon product or part. Beacon shall not be liable for consequential, incidental, special or any other indirect damages resulting from economic loss or property damage sustained by a customer from the use of its products. However, in some states the purchaser may have rights under state law in addition to those provided by this warranty.



Deoxynivalenol (DON) Plate Kit

Cat. # 20-0016

Instructional Booklet

READ COMPLETELY BEFORE USE.

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INTENDED USE

The Beacon Deoxynivalenol (DON) Plate Kit is a competitive ELISA for the quantitative analysis of vomitoxin in wheat, barley, malted barley, corn and oats.

USE PRINCIPLES

The Beacon DON Plate Kit is a competitive enzyme-labeled immunoassay. DON is extracted from a ground sample by shaking with water. The aqueous extract is then filtered and the extract is tested in the immunoassay. DON-HRP enzyme conjugate is pipetted into test wells followed by calibrators or sample extracts and DON Antibody Solution. During the 10-minute incubation period, DON from the sample and DON-HRP enzyme conjugate compete for binding to DON antibody, which is bound to the test well. Following this 10-minute incubation, the contents of the well are removed and the wells are washed to remove any unbound toxin or enzyme-labeled toxin. A clear substrate is then added to the wells and any bound enzyme-toxin conjugate causes the conversion to a blue color. Following a 5-minute incubation, the reaction is stopped and amount of color in each well is read. The color of unknown samples is compared to the color of the calibrators and the DON concentration of the samples is derived.

MATERIALS PROVIDED IN THE BEACON DON PLATE KIT

The kit in its original packaging can be used until the end of the month indicated on the box label when stored at 2 – 8°C.

- 1 plate containing 12 strips of 8 wells coated with goat anti-mouse antibodies
- 5 vials each containing 2 mL of DON calibrators corresponding to 0, 0.2, 0.5, 1.0 and 2.5 µg/mL (ppm) of DON. (Note: Because of the 1:5 dilution of the grain sample in the extraction step, the calibrators actually contain 1/5th of the stated value. No further correction back to the concentration in the original grain sample is required.)
- 1 vial containing 8 mL of DON-HRP Enzyme Conjugate.
- 1 vial containing 14 mL of Substrate.
- 1 vial containing 14 mL of Stop Solution. (Caution! 1N HCl. Handle with care.)
- 1 vial containing 8mL of DON Antibody solution.
- 1 packet Wash solution salts or concentrated liquid.

PERFORMANCE CHARACTERISTICS SPECIFICITY

The antibody utilized in the Beacon DON Kit is specific for deoxynivalenol. The following table shows the relative reactivity for other forms:

Compound	Cross-Reactivity
3-acetyl-deoxynivalenol	<1%
15-acetyl-deoxynivalenol	300%

MATERIALS REQUIRED BUT NOT PROVIDED

1. Laboratory quality distilled or deionized water.
2. Graduated cylinder, 100 mL.
3. Glassware for sample extraction and extract collection.
4. Filter paper
5. Pipet with disposable tips capable of dispensing 50 µL.
6. Multi-channel pipette; 8 channel capable of dispensing 50 and 100 µL or Eppendorf Repeater pipette and tips for dispensing 50 and 100 µL.
7. Paper towels or equivalent absorbent material.
8. Microwell plate or strip reader with 450 nm filter.
9. Timer

PRECAUTIONS

1. Each reagent is optimized for use in the Beacon DON Plate Kit. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other Beacon DON Plate Kits with different Lot numbers.
2. Dilution or adulteration of reagents or samples not called for in the procedure may result in inaccurate results.
3. Do not use reagents after expiration date.
4. Reagents should be brought to room temperature, 20 – 28°C (62 – 82°F) prior to use. Avoid prolonged (> 24 hours) storage at room temperature.
5. Deoxynivalenol is a toxic substance. Dispose of all liquids in a plastic container containing household bleach (minimum 10%). All labware should be soaked for at least 1 hour in a 30% solution of household bleach. Avoid contact of skin and mucous membranes with reagents and sample extracts by wearing gloves and protective apparel. If exposure of skin and mucous membranes to liquids should occur, immediately flush with water.
6. The Stop Solution is 1N hydrochloric acid. Avoid contact with skin and mucous membranes. Immediately clean up any spills and wash area with copious amounts of water. If contact should occur, immediately flush with copious amounts of water.

SAMPLE PREPARATION

1. Grind samples to pass a 20 mesh sieve and thoroughly mix prior to sub-sampling. Samples not being immediately analyzed should be stored refrigerated.
2. Weigh 50 grams of ground sample and combine with 250 mL of laboratory grade water in a clean container with tight fitting lid.
3. Vigorously shake the container for 3 minutes.
4. Allow sample to stand for 2-3 minutes to allow some settling of the slurry.
5. Filter the extract through Whatman #1 filter and collect the extract into a clean container.

WASH SOLUTION PREPARATION

1. Transfer contents of Wash solution packet to 1-liter of lab grade water. Swirl to mix.
2. Fill a wash bottle with Wash Solution.

ASSAY PROCEDURE

(Note: Running calibrators and samples in duplicate will improve assay precision and accuracy.)

1. Allow reagents and sample extracts to reach room temperature prior to running the test.
2. Place the appropriate number of test wells into a microwell holder. Be sure to re-seal unused test wells in the zip-lock bag with desiccant.
3. Dispense **50 µL of Enzyme Conjugate** into each well.
4. Using a pipette with disposable tips, add **50 µL of calibrators and samples** to the appropriate wells. Be sure to use a clean pipet tip for each.
5. Dispense **50 µL of antibody solution** into each well. Swirl plate carefully on bench top to mix contents.

Note: when using more than one 8 well strip in an assay the use of an 8 channel pipette is recommended.

6. Incubate the test wells for **10 minutes**.
7. Dump the contents of the wells into an appropriate waste container. Fill the wells to overflowing with Wash Solution and dump wash. Repeat 4X for a total of five washes.
8. Following the last wash tap the inverted wells onto absorbent paper to remove the last of the wash solution.
9. Dispense **100 µL of Substrate** into each well.
10. Incubate the wells for **5 minutes**.
11. Dispense **100 µL of Stop Solution** into each test well.
12. Read and record the absorbance of the wells at 450nm using a strip or plate reader.