TotalTarget™ Kit for Enogen in corn

Catalog Number AQ-170-BG *Part #13043*

Intended Use

The TotalTarget Kit for Enogen™ Corn is designed for semi-quantitative detection of the amylase protein in Enogen corn in bulk grain samples. Enogen corn derived from transformation Event 3272 contains the transgene *amy797E*, which encodes a synthetic thermostable alpha-amylase protein. The assay will detect the amylase protein found in a sample containing 0.04% Enogen corn (1 positive grain in a 2500 negative grain sample) and requires 10 minutes to run.

Important Notes:

- Before testing, the enclosed Multi-Matrix Barcode Card (MMBC) must be scanned just once for each kit lot to upload information to the QuickScan
- QuickScan Software Version 5.5.1 or later is required

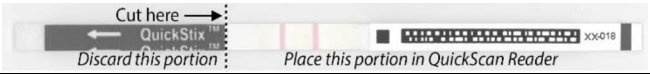
Contents of Kit:

- 50 TotalTarget Strips packed in a moisture-resistant canister
- 50 reaction tubes
- 100 pipette tips (1-200 µL)
- DB6 Buffer
- Multi-Matrix Barcode Card - kit lot specific

How the Test Works

The TotalTarget Kit is a "sandwich" type lateral flow strip test. In the test, bulk grain corn is prepared and tested using a strip coated with antibodies raised against amylase protein. Amylase present in the sample extract binds to the antibodies bound on the strip and is then detected by a second anti-amylase antibody that is gold labeled. A positive sample results in a visual gray line forming about halfway up the strip along with a second control line.

A composite sample is collected, ground, and extracted to solubilize any Enogen present. The extract is further diluted into Buffer before being run on the test strip. Each strip has an absorbent pad at each end. The sample extract travels up the test strip and is absorbed into the larger pad at the top of the strip. At the end of the reaction time, the strip is cut at the top of the arrow tape, the bottom pads are discarded, and the strip is inserted into the QuickScan reader to obtain semi-quantitative results.



Items Not Provided:

- 50 EB17 dissolvable pouches (1 pkt per 25g sample)*
  If not utilizing Common Extraction with TotalTox Aflatoxin
- QuickScan System*
- Incubator (base + block)*
- Bunn grinder or equivalent
- 20-mesh screen (available through Seedburo or other vendor)
- Digital scale for weighing samples
- Extraction cups with lids* or other suitable vessels for sample extraction
- Graduated cylinder*
- Orbital/rotary shaker
- Centrifuge capable of 2000 x *g*, and accessories*
- Pipette to deliver 100 µL*
- Timer
- Scissors
- Distilled, deionized or bottled water

*Available as Accessories

***Available Accessories:**

Item	*Catalog No.*	*Part #*
QuickScan™ System	ACC 331	12721
TotalTox Extraction Set (EB17), 50	ACC 117	12938
5 oz Sample cups/lids *Case of 500; for extracting samples up to 30g*	20-0047	10167
10 oz Sample cups/lids *Case of 100; for extracting samples >30g*	20-0129	12383
Graduated cylinder (100 mL)	ACC 068	11207
MiniPet pipette 100 µL	ACC 041	11202
Centrifugation Set: *Disposables for 50 tests*	ACC 010	11214
Microcentrifuge	ACC 064 E	11204
50g Sample Extraction Set *Additional EB17 dissolvable pouches and sample cups (100)*	ACC 099	12409
DB6 Buffer	KR-268-7	12452
Incubator	ACC BSH301	12458

Sample Preparation

- Collect a composite sample according to your own sampling plan or USDA/FGIS (formerly GIPSA) guidelines. Consult USDA reference documents to help design a plan that fits your needs. Contact Technical Support for more information.
- Grind samples to provide a consistency such that 95% passes through a 20-mesh sieve.
- Mix ground material thoroughly before sub-sampling, to minimize variability.
- Weigh 25 or 50g samples into containers that will allow enough head room for the liquid to move forcefully when shaken vigorously.

Sample Extraction

1. **Extract** sample in buffer. Use EB17 provided in TotalTox Aflatoxin kit, or purchase separately. Use distilled, deionized, or flat (non-carbonated) bottled water. If testing 50-gram samples, additional EB17 Buffer pouches and larger extraction vessels are required (50g Sample Set, order Catalog No. ACC-099).

25g samples	***50 g samples***
• Add 1 EB17 pouch to sample • Then add 75 mL water	• Add 2 EB17 pouches to sample • Then add 150 mL water
Wet sample immediately, by vigorously shaking for 10 seconds by hand. If needed, shake the sample against the palm of your other hand or a hard surface to loosen up any dry sample areas. **Immediately** proceed to next shaking step.	

2. **Shake** sample, mechanically or by hand.

Shaker Table: mix at highest speed (≥ 300 rpm) for 1 minute	***By Hand:*** shake vigorously for 2 minutes

3. **Clarify** sample: Immediately fill a microcentrifuge tube with extract, avoiding the top foamy layer if present, and centrifuge it for 30 seconds at 2000 x *g* (not RPM). Consult centrifuge manual for *g* force calculation, and follow manufacturer's instructions for operation and balancing. Use the top layer of extract; there may be a solid floating particulate layer above the extract that should not be used for testing

TIPS!

Get Complete Extraction
- Fully wet samples before the next shaking step
- Avoid delay between buffer addition and shaking
- Assure liquid is moving forcefully though the sample while shaking

For Best Performance
- Pipette up and down while mixing
- Read strips promptly after run time

Avoid Contamination
- Use a new Reaction Tube per test
- Keep DB6 capped, when possible
- Use new pipette tips for each step

How to Run the TotalTarget Test

- Review Sample Preparation above for grinding consistency and notes.
- Turn on the incubator and set to 22°C for a minimum of 10 minutes before testing. Ensure that the temperature display has stabilized and indicates "OK" before starting the assay. All reagents should be at room temperature.

1. Add 100 µL DB6 to the Reaction Tube (discard tip). Please note: if running alongside TotalTox mycotoxin test kits, label test tubes and be sure to use the specified Dilution Buffer for each test (for example: TotalTox Aflatoxin uses DB5).
2. Add 100 µL clarified extract to the Reaction Tube
3. Mix thoroughly with extract pipette tip, discard tip
4. Place the Reaction Tube in the 22°C incubator; equilibrate for 2 minutes. Note: tube acclimation step is only required if the temperature of the testing environment is unknown or outside of 20 - 24°C (68 - 75°F)
5. Add test strip to tube, arrows down, wait 10 minutes (run time)
6. Immediately cut strips at the top of the arrow tape (discard bottom pads)
7. Insert strip into QuickScan Reader and click Read Test
8. QuickScan will return a result that is either "Negative" (<0.04%) or "Positive" (≥0.04%)

Use of the QuickScan System

Detailed instructions for use of the QuickScan System are supplied with each unit and can also be found at www.envirologix.com/support. The lot-specific Multi-Matrix Barcode Card (MMBC) must be scanned into the system prior to testing. In summary, a strip is inserted into the reader and the strips are read by touching or clicking on the "Read Test" area of the screen. The "Select Matrix Groups" screen will appear if more than one barcode was scanned into the system from the MMBC. Select the group that displays the matrix run. Results are then recorded in an electronic worksheet, allowing each user to report and track data easily.

Kit Storage

TotalTarget Enogen strips can be stored in their tightly capped canister at room temperature, or refrigerated for a longer shelf life. Note the shelf life on the kit box for each storage temperature. **NOTE: The DB6 buffer must be stored refrigerated.** The kit may be used in field applications; however, prolonged exposure to high temperatures may adversely affect the test results. Do not open the desiccated canister until ready to use the test strips.

Notes

- This product is currently not applicable for use in testing any other crops beyond those specified in this Product Insert.
- This kit is designed to give a result using the QuickScan System and is not intended to be visually interpreted.
- The assay has been optimized to be used with the protocol provided in the kit. Deviation from this protocol may invalidate the results of the test.
- As with all tests, it is recommended that results be confirmed by an alternate method if necessary.
- Immediate shaking of the sample after water addition is critical to ensure the EB17 packet does not cause clumps which may interfere with test results.
- Pipettes lose calibration accuracy over time. Calibrate or replace pipettes at least annually.
- The results generated through the proper use of this diagnostic tool reflect the condition of the working sample directly tested. Extrapolation as to the condition of the originating lot, from which the working sample was derived, should be based on sound sampling procedures and statistical calculations which address random sampling effects, non-random seed lot sampling effects and assay system uncertainty. A negative result obtained when properly testing the working sample does not necessarily mean the originating lot is entirely negative for the analyte or protein in question.
- DO NOT leave in direct sunlight or in vehicle. Protect all components from hot or cold extremes of temperature when not in use.



For Technical Support Contact Us At:

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www.envirologix.com

LIMITED WARRANTY

EnviroLogix Inc. ("EnviroLogix") warrants the products sold hereunder ("the Products") 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. If the Products do not conform to this Limited Warranty and the customer notifies EnviroLogix in writing of such defects during the warranty period, including an offer by the customer to return the Products to EnviroLogix for evaluation, EnviroLogix will repair or replace, at its option, any product or part thereof that proves defective in materials or workmanship within the warranty period.

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This Limited Warranty states the entire obligation of EnviroLogix with respect to the Products. If any part of this Limited Warranty is determined to be void or illegal, the remainder shall remain in full force and effect.

License

EnviroLogix has developed this kit using proprietary reagents.

This test kit has been approved by Syngenta for detection of the amylase protein in Enogen™ corn.

Enogen™ is a trademark of a Syngenta Group Company
EnviroLogix, the EnviroLogix logo, QuickScan, and TotalTarget are trademarks of EnviroLogix Inc.

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Safety data sheet

SECTION 1. Identification of the substance/mixture and of the company/undertaking

1.1 Product identifier
Trade name: **DB 6 Dilution Buffer**
Part number 11151 (KR-268)

1.2 Relevant identified uses of the substance or mixture and uses advised against application of the substance / the preparation : Laboratory chemicals; kit component. Not to be used for purposes other than those specified in product literature.

1.3 Details of the supplier of the safety data sheet
Manufacturer/Supplier: EnviroLogix Inc., 500 Riverside Industrial Pkwy, Portland ME 04103, USA
Phone: (207) 797-0300

1.4 Emergency telephone number: (207) 797-0300 Technical Service

SECTION 2. Hazards identification.

2.1 Classification of the substance or mixture

Classification according to 29CFR 1910.1200: Not Classified

2.2 Label elements
Labeling according to 29CFR 1910.1200

Pictogram: None
Signal word: None
Hazard Statements: None

2.3 Other Statements: None

SECTION 3. Composition/information on ingredients.

3.2 Mixture

Chemical name	CAS No	EC No	Classification According to 29CFR 1910.1200	Amount (%)
Sodium Tetraborate Decahydrate	1303-96-4	215-540-4	H360 Rep 1B	< 3 %

SECTION 4. First aid measures.

4.1 Description of first aid measures

After inhalation : In case of inhalation. Remove to fresh air. If not breathing give artificial respiration. Get medical attention immediately.

After skin contact : In case of skin contact. Remove contaminated clothing and shoes immediately. Wash affected area with mild soap or detergent for at least 10 minutes or until no evidence of chemical remains.

After eye contact : In case of eye contact, immediately flush eyes with plenty of water for at least 15 minutes. Lifting eyelids occasionally, until no evidence of chemical remains. Get medical attention immediately.

After swallowing : In case of ingestion. DO NOT Induce vomiting unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. Call a physician immediately.

4.2 Most important symptoms and effects, both acute and delayed: None

4.3 Indication of any immediate medical attention and special treatment needed: None

SECTION 5. Firefighting measures.

5.1 Extinguishing media: CO_2, extinguishing powder or water spray. Fight larger fires with water spray or alcohol resistant foam.

5.2 Special hazards arising from the substance or mixture: None

5.3 Advice for firefighters: Wear protective gear appropriate for fire conditions including respiratory protective gear.

SECTION 6. Accidental release measures.

6.1 Personal precautions, protective equipment and emergency procedures: In the case of spilled mixture wear gloves to prevent skin contact. In the case of a large spill, additional protection is recommended.

6.2 Environmental precautions: Do not discharge mixture to sewer system or waterways.

6.3 Methods and material for containment and cleanup: Absorb in paper towel or suitable absorbent for larger spills and discard in appropriate waste. Clean with water afterwards.

6.4 References to other sections: For safe handling refer to Section 7. For information on PPE refer to Section 8. For disposal refer to Section 13

SECTION 7. Handling and storage.

7.1 Precautions for safe handling: Practice good chemical hygiene when handling. Avoid contact with eyes, skin, and clothing.

7.2 Conditions for safe storage, including any incompatibilities: Store in tightly closed, non-metal container, in a corrosive compatible area. Prevent direct sunlight and heat. Store in well aired storage rooms.

7.3 Specific end use(s): Apart from the uses mentioned in section 1.2, no other specific uses are stipulated

SECTION 8. Exposure controls/personal protection.

8.1 Exposure limits:
Components with limit values that require monitoring at the workplace:

	EH40/2005	OSHA
Sodium Tetraborate Decahydrate	8 Hr TWA = 5mg/m^3	8 Hr TWA = 10 mg/m^3

8.2 Exposure Controls:

8.2.1 Engineering controls: Facilities using this mixture should be equipped with an eyewash and safety shower. Use general or local exhaust ventilation to keep airborne concentrations below permissible exposure limits.

8.2.2 General protective and hygienic measures: The usual precautionary measures should be adhered to when handling chemicals.

Eye Protection: Safety glasses with side shields, goggles. Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166 (EU). Eye and face protection regulations are described by OSHA (US) in 29CFR1910.133. Do not wear contact lenses when working with chemicals

Hand Protection: Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands. The selected protective gloves have to satisfy the specifications of EU Directive 89/686/EEC and the standard EN 374 derived from it.

Breathing Equipment: Appropriate respiratory protection should be determined according to local conditions using risk analysis protocols. An approved disposable air purifying particulate respirator may be used as a backup to engineering controls. Always use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

8.2.3 Environmental exposure controls: Contain spills, do not allow into environment

SECTION 9. Physical and chemical properties.

9.1 Information on basic physical and chemical properties:

a) Appearance: Clear liquid, colorless to slight yellow.
b) Odor: None
c) Odor Threshold: No Data Available
d) pH: 8.6
e) Melting point/freezing point: No Data Available
f) Boiling point/Boiling range: No Data Available.
g) Flash point: Not applicable.
h) Evaporation rate: No Data Available
i) Flammability (solid, gaseous): No Data Available
j) Upper/lower flammability or explosive limits: No Data Available
k) Vapor pressure: No Data Available
l) Vapor density No Data Available
m) Relative density: No Data Available
n) Solubility(ies): Fully miscible, water.
o) Partition Coefficient: n-Octanol/water: No Data Available
p) Auto-ignition temperature: No Data Available
q) Decomposition temperature: No Data Available
r) Viscosity: No Data Available
s) Explosive properties: No Data Available.
t) Oxidizing properties: No Data Available

9.2 Other information: No further relevant information available.

SECTION 10. Stability and reactivity.

10.1 Reactivity: No data available

10.2 Chemical Stability: Stable under normal temperatures and pressures.

10.3 Possibility of hazardous reactions: Under normal conditions of storage and use, hazardous reactions will not occur.

10 4 Conditions to avoid: No specific data

10.5 Incompatible materials: No Data Available.

10.6 Hazardous decomposition products: Under normal conditions of storage and use, hazardous decompositions products should not be produced.

SECTION 11. Toxicological information.

Information on Toxicological Effects

Acute effects (toxicity tests): No Data Available

Sensitization: No sensitizing effects known

CMR (carcinogenicity, mutagenicity and toxicity for reproduction) effects: No CMR effects.

Additional toxicological information: No Additional Information

SECTION 12. Ecological information.

12.1 Toxicity: No Data Available

12.2 Persistence and degradability : No Data Available

12.3 Bio accumulative potential: No Data Available

12.4 Mobility in soil : No Data Available

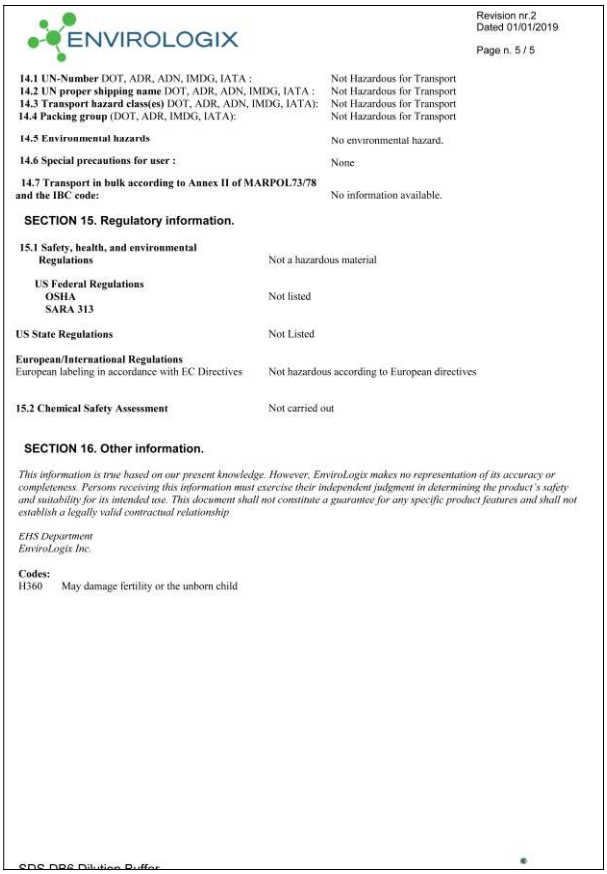
12.5 Results of PBT and vPvB assessment: Not available as a chemical safety assessment, not required/not conducted.

12.6 Other adverse effects: No Data Available

SECTION 13. Disposal considerations.

Waste treatment methods: Contact a licensed professional waste disposal service to dispose of this material. Disposal of surplus or waste solutions must be in accordance with applicable local, state, and national laws and regulations.

SECTION 14. Transport information.



ENVIROLOGIX

Revision nr.2
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Page n. 5 / 5

14.1 UN-Number DOT, ADR, ADN, IMDG, IATA :	Not Hazardous for Transport
14.2 UN proper shipping name DOT, ADR, ADN, IMDG, IATA :	Not Hazardous for Transport
14.3 Transport hazard class(es) DOT, ADR, ADN, IMDG, IATA):	Not Hazardous for Transport
14.4 Packing group (DOT, ADR, IMDG, IATA):	Not Hazardous for Transport
14.5 Environmental hazards	No environmental hazard.
14.6 Special precautions for user :	None
14.7 Transport in bulk according to Annex II of MARPOL73/78 and the IBC code:	No information available.

SECTION 15. Regulatory information.

15.1 Safety, health, and environmental Regulations	Not a hazardous material
US Federal Regulations	
OSHA	Not listed
SARA 313	
US State Regulations	Not Listed
European/International Regulations	
European labeling in accordance with EC Directives	Not hazardous according to European directives
15.2 Chemical Safety Assessment	Not carried out

SECTION 16. Other information.

This information is true based on our present knowledge. However, EnviroLogix makes no representation of its accuracy or completeness. Persons receiving this information must exercise their independent judgment in determining the product's safety and suitability for its intended use. This document shall not constitute a guarantee for any specific product features and shall not establish a legally valid contractual relationship

EHS Department
EnviroLogix Inc.

Codes:
H360 May damage fertility or the unborn child

SDS DB6 Dilution Buffer

Summary Guide for Testing TotalTarget Enogen								
Approved Matrix	**Add to Sample Extraction Vessel (in this order)**	**Then shake immediately**	**Clarify**	**Run the Base Range Protocol**	**Pre-mix as noted, then Transfer to Reaction Tube**	**Add Reaction Tube to Incubator Set at 22°C**	**Add Strip for**	**Read in QuickScan: Dilution Tab on the Result Page Should Display**
TotalTarget Enogen Corn (MG1)	1. 25g sample 2. 1 EB17 pouch* 3. 75 mL water† 4. Immediately shake vigorously for 10 seconds by hand ---------*OR*--------- 1. 50g sample 2. 2 EB17 pouches* 3. 150 mL water† 4. Immediately shake vigorously for 10 seconds by hand	1 min highest speed on shaker table or 2 min by hand	Centrifuge 30 sec. at 2000 x *g*	Base Range Protocol	**Pre-Mix** 100 µL **DB6**‡ buffer + 100 µL clarified extract in Reaction Tube	Acclimate tube for 2 min^	10 min.	1:1 (this is software default)

Notes:

*Use EB17 pouches from TotalTox Aflatoxin Kit; otherwise purchase ACC-117 TotalTox Extraction Set

†Use distilled, deionized, or flat (non-carbonated) bottled water

‡ Use the Dilution Buffer provided with each kit; and label tubes to avoid using the wrong DB when testing

^The tube acclimation step is only required if the temperature of the testing environment is unknown or outside of 20 - 24°C (68 - 75°F)

<table>
<tr><th colspan="8">Summary Guide for Coordinated Testing with TotalTox Common Extraction</th></tr>
<tr><th>Approved Matrix</th><th>Add to Sample Extraction Vessel (in this order)</th><th>Then shake immediately</th><th>Clarify</th><th>Run the Base Range Protocol</th><th>Pre-mix as noted, then Transfer to Reaction Tube</th><th>Add Reaction Tube to Incubator Set at 22°C</th><th>Add Strip for</th><th>Read in QuickScan: Dilution Tab on the Result Page Should Display</th></tr>
<tr><td>TotalTarget Enogen

Corn (MG1)</td><td rowspan="2">1. 25g sample
2. 1 EB17 pouch*
3. 75 mL water†
4. Immediately shake vigorously for 10 seconds by hand
---------OR---------
1. 50g sample
2. 2 EB17 pouches*
3. 150 mL water†
4. Immediately shake vigorously for 10 seconds by hand</td><td rowspan="2">1 min highest speed on shaker table
<u>or</u>
2 min by hand</td><td rowspan="2">Centrifuge 30 sec. at 2000 x g</td><td rowspan="2">Base Range Protocol</td><td>In Tube A:
<u>Pre-Mix</u>
100 µL DB6‡ buffer + 100 µL clarified extract in Reaction Tube, Mix Well</td><td rowspan="2">Acclimate tube for 2 min^</td><td>10 min.</td><td>1:1 (this is software default)</td></tr>
<tr><td>TotalTarget Enogen Corn

alongside

TotalTox

Corn Common Extraction for Mycotoxins</td><td>In Tube B:
<u>Pre-Mix</u>
100 µL DB5‡ buffer + 100 µL clarified extract in Reaction Tube, Mix Well</td><td>4 min.</td><td>1:1 (this is software default)

If results are above the stated base range, follow TotalTox instructions for extended dilution & quantification</td></tr>
</table>

<u>Notes</u>:

*Use EB17 pouches from TotalTox Aflatoxin Kit; otherwise purchase ACC-117 TotalTox Extraction Set

†Use distilled, deionized, or flat (non-carbonated) bottled water

‡ Use the Dilution Buffer provided with each kit; and label tubes to avoid using the wrong DB when testing

^The tube acclimation step is only required if the temperature of the testing environment is unknown or outside of 20 - 24°C (68 - 75°F)