

Catalog No. ACC-091

Part #12038

Set Contains:

- MB9 Extraction Buffer
- D4 Dilution Buffer
- 2.0 mL clear micro-centrifuge tubes (25) for extraction
- 1.5 mL blue micro-centrifuge tubes (25) for dilution
- Grain scoops (4)

Materials Not Provided:

- Precision pipette(s) capable of delivering 25-1000 μ L
- Pipette tips
- Dry heat block capable of $95 \pm 1^\circ\text{C}$, with insert suitable for 2 mL tubes
- Vortex
- Micro-centrifuge capable of 10,000 x g
- Timer

Intended Use

This Set provides for extraction for the detection of DNA from ground soybean, soybean meal, and soybean flakes when used in combination with the following DNABLE Kits:

	Catalog No.	Part #
Multi-Trait (RR1, RR2, LL)	DF-031	11979
Multi-Trait (RR1, RR2, LL, DMO)	DF-041	12241
<i>dmo</i> (DMO)	DF-050	12242
<i>pat / pat</i> (LL)	DF-014	11978
<i>cp4 epsps</i> (RR1 and RR2)	DF-012	11977
<i>cp4 epsps</i> (RR1, GTS 40-3-2)	DF-112	11976
<i>cp4 epsps</i> (RR2, MON89788)	DF-212	12129

Contact Technical Support for specific recommendations on other matrix types.

Intended User

DNABLE is designed to be simple and user friendly. It is designed for use by personnel with appropriate training in Molecular Assay techniques. Training

specific to the DNABLE assay will be provided by EnviroLogix; contact Technical Service or visit envirolgix.com for more information.

How the Kit Works

An aliquot of MB9 buffer is added to a micro-centrifuge tube followed by ground soybean, soybean meal, or soybean flakes. The sample is heated to enable the extraction of DNA. A centrifugation and dilution step follows.

Precautions and Notes

DNABLE is a highly sensitive assay. Therefore the following precautions are recommended to reduce the chance of sample contamination:

- Separated work areas are recommended for each of the following:
 - DNABLE sample preparation
 - DNABLE amplification and detection
- Clean the work station and pipettes with 10% bleach before and after use
- Do not reuse kit disposables
- Grain scoops are reusable: wash in 10% bleach and ensure the scoop is dry before reusing
- Change pipette tips in between samples, including replicates from the same sample extract
- Wear gloves and change between handling of samples
- Avoid delays between sample preparation steps and between sample preparation and DNA amplification
- MB9 is stable for 1 year post manufacture when stored refrigerated (2-8°C)

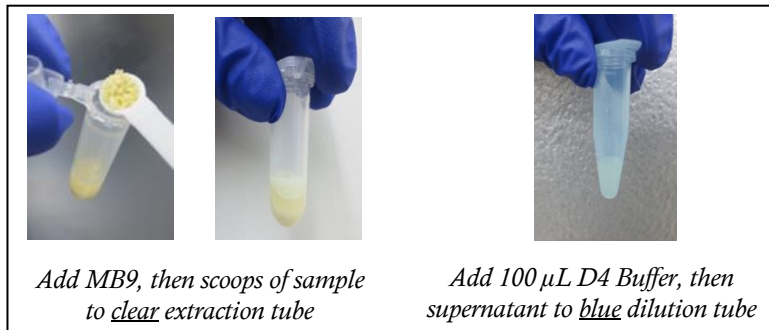
Sample Preparation and Extraction

1. Pre-heat a dry heat block
 - 95°C for bulk soybeans or soybean meal, or
 - 85°C for soybean flakes

Allow heat block to warm for 30 minutes. Verify heat block is holding temperature with $\pm 1.5^\circ\text{C}$ using a simple thermometer.

2. Choose protocol based on sample type:

<p>a. Soybeans</p> <ol style="list-style-type: none"> Grind a minimum of 200 soybeans (weight to be determined by average seed weight) in an Oster blender for 20 seconds. Repeat 2 additional times for total of 3 grinds. IMPORTANT: Shake MB9 bottle for 5 seconds before use. This is to ensure that any undissolved particulate matter is homogenously distributed in the buffer. Add 600 μL MB9 Extraction Buffer to 2.0 mL clear extraction tube (1 tube per sample). Using a clean grain scoop*, add two packed level scoops of ground soybean to extraction tube with buffer, then cap tightly. Flick tube to ensure complete suspension of sample. *use a clean scoop for each grind to avoid contamination of samples – see instructions in Precautions & Notes Heat tube from previous step at 95°C for 6 minutes (± 30 seconds). Remove the 2 mL extraction tube from heat block and vortex 5 seconds to mix. Spin sample at 10,000 x g for 3 minutes (± 30 seconds). Add 100 μL of D4 Buffer to a clean 1.5 mL blue dilution tube. Add 100 μL of supernatant from spun sample, taking care to avoid settled soybean. Vortex to mix. 25 μL of this diluted crude extract will be used in the subsequent DNABLE reaction (DNABLE Kit, Sample Preparation Step 3). 	<p>b. Soybean meal and soybean flakes</p> <ol style="list-style-type: none"> Grind a representative sample of soybean meal for 20 seconds in an Oster blender. IMPORTANT: Shake MB9 bottle for 5 seconds before use. This is to ensure that any undissolved particulate matter is homogenously distributed in the buffer. Add 1 mL MB9 Extraction Buffer to a 2.0 mL clear extraction tube (1 tube per sample). Using a clean grain scoop*, add four (4) unpacked level scoops of soybean meal to the extraction tube with buffer, cap tightly, and shake to mix, ensuring that there is no bubble at the bottom of the tube. *use a clean scoop for each grind to avoid contamination of samples – see instructions in Precautions & Notes Heat tube from previous step at 95°C (soybean meal) or 85°C (soybean flakes) for 6 minutes (± 30 seconds). Remove the 2 mL extraction tube from heat block and vortex 5 seconds to mix. Spin sample at 10,000 x g for 3 minutes (± 30 seconds). Add 100 μL of D4 Buffer to a clean 1.5 mL blue dilution tube. Remove 100 μL of supernatant from the center portion of the spun sample (avoiding settled material as well as top layer) and add to tube. Vortex to mix. 25 μL of this diluted crude extract will be used in the DNABLE reaction (DNABLE Kit, Sample Preparation Step 3).
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