

## Highlights:

- Molecular detection of *Mycoplasma synoviae* in poultry tracheal swabs
- Rapid amplification and detection in 15 minute assay

## Contents of DNABLE Kit:

- A. RB1 Reaction Buffer
- B. *M. synoviae* Master Mix
- C. Flat Caps
- D. MB1 Extraction Buffer
- E. 2.0 mL Extraction Tubes
- F. Pipette tips

## Materials Not Provided:

- Pipettes (for 600 µL)
- 8-well AmpliFire Reader\*

\*available through EnviroLogix

Catalog No. DF-030 MS

Part #11781

## Intended Use

This test kit is intended for qualitative detection of DNA from *Mycoplasma synoviae* (MS) in tracheal swabs. The results of this test may facilitate rapid, point of need detection of MS to manage of poultry flock health.

## How the Test Works

DNABLE is an isothermal nucleic acid amplification technology enabling rapid amplification of a specific DNA target. In this test, samples are collected, processed, and added to the reaction buffer. The reaction buffer containing sample is then transferred to the lyophilized master mix, containing all the reagents needed to specifically recognize, amplify and detect the MS specific DNA.

The amplified MS-specific DNA is detected in real-time and the results are displayed and interpreted within 15 minutes using our 8-well DNABLE Reader.

## Precautions and Notes

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

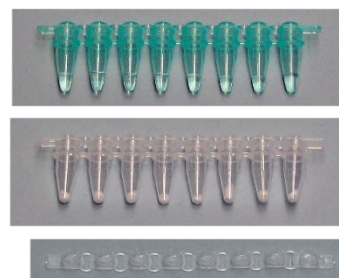
- Clean the work stations and pipettes before and after use with 10% bleach solution
- It is recommended to physically separate sample preparation activities from DNABLE assay activity
- Do not reuse kit disposables

- Use fresh pipette tips for each sample
- Discard used tips in a sealed container containing 10% bleach solution
- Use careful pipetting techniques to avoid cross-contamination between samples; avoid reaching over or pipetting over open tubes
- Wear disposable gloves when handling of samples

**Important:** Never open reaction tubes after reaction has occurred, as this will release amplified material into the environment and may contaminate subsequent reactions. Care should be taken when disposing of run reaction tubes to avoid possibility of tube leakage. Place completed reaction tubes back in original zippered pouch prior to disposal.

## Kit Components

- A. RB1 Reaction Buffer: Provided in green 8-well strip tubes (6)
- B. *M. synoviae* Master Mix: Lyophilized reagents provided in clear 8-well strip tubes (6 strips)
- C. Flat Caps: used for capping the clear tubes prior to assay start (6 strips)
- D. MB1 Extraction Buffer: Two 20 mL bottles of extraction buffer (for sample preparation)
- E. 2.0 mL Extraction Tubes (50): Two bags of 25 tubes for sample extraction



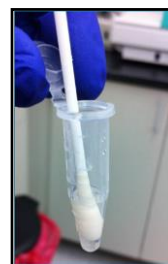
## Before Testing

- Remove needed DNable Kit reagents from refrigerated storage. Allow reagents to come to room temperature before opening sealed white pouches.
- Turn on the 8-well AmpliFire Reader using power button on the right side of the instrument.
- Ensure that all assay reagents, extracted sample, pipettes and flat caps are ready for use.



## Tracheal Swab Sample Collection and Preparation

- Three tracheal swabs may be pooled per sample.
- For each sample, use a clean pipette tip to add **600 µL of MB1 Extraction Buffer** to each extraction tube.
- Place the tracheal swab into the extraction buffer and swirl** around the tube wall, then remove the swab.
- Repeat step 3** with the remaining two (2) tracheal swabs in this pooled sample.
- Vortex the extraction tube for 5 seconds** to homogenize the sample.
- Remove green Reaction Buffer strip tubes from the kit. Mark the left end tube to note orientation.



10 µL extract per well  
(each pooled sample = 1 well)



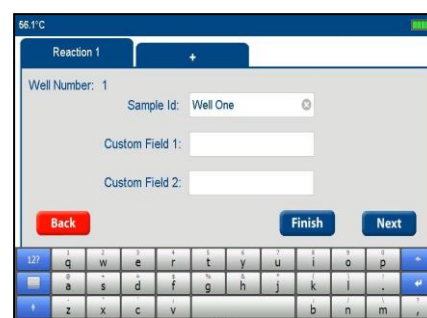
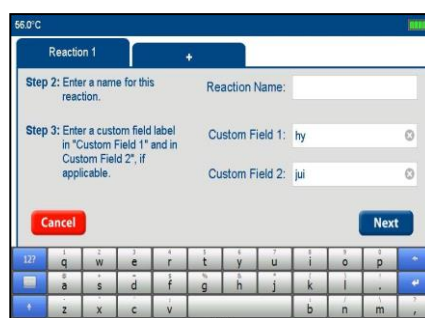
**Important:** Tap down on green strip tube to ensure that the entire buffer volume is at the bottom of the tubes prior to opening.

- Pipette **10 µL of sample extract** to a single well of the reaction buffer 8-well strip tube. Pipette the 10 µL volume directly into the reaction buffer. Repeat for wells 2-8, using a fresh pipette tip for each sample.
- Recap tubes and tap down to ensure all liquid is at the bottom of the tube.

## How to Run the DNable Assay

### DNable assay protocol

- On the AmpliFire screen, select **“Execute Reaction”** then **“Scan Barcode”**. Use the barcode on the master mix foil pouch to scan the MS protocol on the 8-well AmpliFire Reader. MS\_Lot # will display. Select **“Next”**.
- Under **“Reaction Name”** enter an appropriate reaction description. **This description is placed at the beginning of the file name.** Select **“Next”**.
- To enter sample specific information, add sample descriptions to the screens for Wells 1 through 8, clicking **“Next”** to advance to each Well. Select **“Finish”** to skip well-specific sample entry.



- Remove clear Master Mix tubes from the foil pouch and gently tap down to ensure that the white pellet is at the bottom of the tubes.

**Important:** Mark flat cap for orientation of the clear Master Mix tubes (writing on clear tubes will interfere with results interpretation or leave marker residue in instrument).

- Using a multichannel pipette, transfer **50  $\mu$ L from green strip tubes (containing sample)** to clear Master Mix tubes. Do **not** mix within the clear tube.

- Cap** Master Mix tubes with provided **Flat Caps** strip.

**Important:** Ensure that the tubes are **completely sealed** with flat caps

- Gently flick down on the resuspended, capped master mix to ensure that no bubbles are at the bottom of the tube and that master mix is fully resuspended.



- Inspect tube to ensure that **no air bubbles are present within the sample volume** (a bubble at the top is fine) and that **cap is completely sealed**.



- When the strip is ready select **“Start”**. Place resuspended, capped clear strip tube in instrument and press **“Ok”**.
- After 15 minutes, the Amplifire will produce a short beeping sound and display final results. Results will be interpreted as Not Detected (-) or Positive (+).

**Important:** Positive results may be interpreted prior to assay completion, but the full assay time must be complete for complete result interpretation. (Empty wells will be interpreted as negative.)

- After the assay is complete, carefully **remove run reaction strip tubes from instrument and place in opened foil pouch** (used to store master mix), seal and discard in waste container.
- To export results, return to the home screen, then **“View Results”**. Insert a USB storage device into instrument (left side) and select each run to export and **“Export Selected”** and **“OK.”** The results will be saved in a PDF summary report as well as .csv file format.

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For Technical Support  
Contact Us At:

**EnviroLogix Inc.**  
500 Riverside Industrial Parkway  
Portland, ME 04103-1486 USA  
**Tel: (207) 797-0300**  
**Toll Free: 866-408-4597**  
**Fax: (207) 797-7533**

e-mail: [dnable@envirolgix.com](mailto:dnable@envirolgix.com)

Website: [www.envirolgix.com](http://www.envirolgix.com)

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