

Directions for Use of MicroSnap TOTAL (Total Microbial Activity Test)



Catalog Number: MS -TOTAL

Parts Required:

1. MicroSnap TOTAL Enrichment Swab Device, Part # MS-TES100
2. MicroSnap TOTAL Detection Device, Part # MS-TOTAL100

Description/ Intended Use:

MicroSnap TOTAL (Total Microbial Activity Test) is a rapid bioluminescent method for the detection and enumeration of total population of bacteria or total contamination in a sample providing results in less than 8 hours. MicroSnap TOTAL consists of an Enrichment Swab Device containing proprietary growth media and a Detection Device containing bioluminescent reagents in which biomarkers produced by bacteria are measured using a small portable luminometer.

The two step test procedure requires a short incubation period allowing the growth of bacteria followed by the detection step. During incubation in the Enrichment media, the number of bacteria is increased and potential sample interference is reduced. As the bacteria grow they use up the available food resources in the media and generate enzymatic biomarkers. The greater the number of bacteria in the sample; the higher the biomarker concentration is after a set time interval. An aliquot of the Enriched sample is transferred to the Detection device, activated, mixed and measured using a luminometer. The light output is directly proportional to the concentration of biomarkers and number of bacteria present.

MicroSnap TOTAL can be used to test environmental surfaces, product samples, water and other filterable liquids.

Material and Reagents required for food samples but not provided:

Sample preparation diluents, sample bags and homogenizing equipment

Recommended Equipment:

- o Incubator set at 30°C ± 1°C
- o Compatible luminometers
 - EnSURE
 - SystemSURE Plus

Test Procedure: Refer to diagrams on the next page.

Step 1: Enrichment: Sample, Activate and Incubate.

1. **Collect sample** and place in the MicroSnap TOTAL Enrichment device (Part. # MS- TES100)
Samples can be of the following types;
 - i. Surface: Swab the area of interest (typically 4 x 4 inches; 10 x 10 cm).
 - ii. 1mL liquid food, beverage or water: samples added directly to Enrichment device.
 - iii. 1mL 10% w/v (weight / volume) food homogenate: added directly to Enrichment device. The food homogenate is prepared using standard microbiological procedures. For unknown sample contamination rates or for better resolution dilutions below 10% should be performed, see section Dilutions.
2. Re-insert the swab into swab tube.
3. **Activate** device by bending the bulb to break the Snap Valve pin at the base.
4. After the Snap Valve pin is broken, separate the bulb / swab assembly (about 1 – 2") from the swab tube to release the internal pressure and squeeze the bulb 2 times to release the Enrichment Media into swab tube. Ensure that most of the Enrichment Media is in the bottom of the swab tube. Reinsert the bulb / swab assembly firmly into the swab tube to seal the device.
5. Shake the tube gently to mix sample and Enrichment Media.
6. **Incubate** at 30° ± 1°C for 7 hours. This will be referred to as the **Enriched Sample**.

Step 2: Detection: Transfer Enriched Sample to detection device, Activate, Mix and Measure.

1. Allow the MicroSnap TOTAL Detection Device (MS-TOTAL100) to equilibrate to room temperature (10 minutes). Shake the test device by tapping on the palm of your hand 5 times (to bring the condensation in the tube to the bottom of the tube prior to adding the Enriched Sample to the tube. This will facilitate the mixing of the Enriched Sample with the solution in the tube.
2. **Aseptically transfer an aliquot of the Enriched sample** from the Enrichment device (MS-TES100) to the MicroSnap TOTAL Detection Device (MS-TOTAL100). Optimum volume is 0.1ml, (3 drops). A simple and easy way to transfer sample from the Enrichment device to the Detection device is to squeeze the bulb of the Enrichment Swab sucking the sample back up into the Snap Valve bulb. Then separate the bulb and tube of the Enrichment device. Squeeze the bulb, carefully dispensing 3 drops (~ 0.1ml) into the Detection device. Re-insert Snap valve bulb in the Enrichment and Detection device filling to the marked fill line is exactly 0.1ml. Excess fill does not affect the test.
3. **Activate** Detection Device (MS-TOTAL100). Bend the bulb to break the Snap Valve *pin*. Squeeze bulb 3 times to release the reagent..
4. Shake gently to **mix**.
5. Insert the whole device into the luminometer; close the lid and holding the unit upright, press "OK" button to initiate the measurement. Results will appear after the 15 second count down.
6. **Read** result. Interpret the result as directed below.

Interpretation of Results:

The results on the luminometer are displayed as Relative Light Units (RLU). The RLU output is proportional to the starting inoculum and the corresponding bacteria equivalent numbers (expressed as Colony Forming Units, CFU). Table 1 shows the equivalent CFU values for RLU measurements obtained using the 2 different Hygiena luminometers.

Table 1

Correlation between CFU/ mL and average RLU
After 7 hour Incubation at 30°C

CFU per ml/100cm ²	EnSURE
< 100	< 10
< 200	< 20
< 300	< 30
< 500	< 50
< 1,000	< 100
< 10,000	< 1,000
< 100,000	< 9,999

Controls

It is advisable to run positive and negative controls according to Good Laboratory Practices.

Dilutions

The use of dilutions for samples is advised when the total number of bacteria is unknown or the total number of bacteria may be increasing due to process changes.

Diluents can be used to dilute the initial 10% homogenate further to 1%, 0.1% and 0.01% if required. On running the assay with more than 1 dilution, back calculations should be performed on the transformed RLU to CFU prior to multiplying by the dilution factor to obtain the CFU/g.

Alternatively, use the online TVC calculator available online or download from the Hygiena website.

Safety & Precautions:

Components of MicroSnap TOTAL Enrichment device (MS-TES100) and Detection device (MS-TOTAL100) do not pose any health risk when used correctly. Used devices that confirm positive results may be bio- hazardous and should be disposed of safely in compliance with Good Laboratory Practice and Health and Safety Regulations.

Disinfect before disposal. MicroSnap devices can be disinfected by autoclaving or by soaking in 20% bleach for 1 hour. Then, they can be placed in the trash. Alternatively, they can be taken to a biohazardous disposal facility.

1. MS-TOTAL100 Device is designed for a single use. Do not reuse.
2. Do not use devices after Expiration Date.
3. Sampling should be done aseptically, to avoid cross contamination.
4. Ensure proper incubation temperature and time for the test application.

Storage & Shelf Life:

Boxes and bags of devices must be stored at 2° - 8°C.

Devices have a shelf life of 12 months. Check expiration date on label.

Hygiena Liability: As with any culture medium, MicroSnap TOTAL results do not constitute a guarantee of quality of food, beverage products or processes that are tested with these devices. Hygiena will not be liable to user or others for any loss or damage, whether direct or indirect, incidental or consequential from use of this device. If this product is proven to be defective, Hygiena's sole obligation will be to replace product, or at its discretion, refund the purchase price. Promptly notify Hygiena within 5 days of discovery of any suspected defect and return product to Hygiena. Please call Customer Service for a Returned Goods Authorization Number.

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MicroSnap™ Total

Step 1: Enrichment of Environmental Surface Swab, Liquid and Solid Samples

1 **Surface**

i. **Surface:** Swab a 10x10cm area or larger depending on protocol with the **MicroSnap TOTAL Enrichment device (MS-TES100)**.

OR

ii. Liquids: 1ml liquid food, beverage or water sample added directly to **MicroSnap TOTAL Enrichment device (MS-TES100)**.

OR

iii. Solid Samples: 1ml 10% w/v suspension of solid samples added directly to **MicroSnap TOTAL Enrichment device (MS-TES100)**.

2

2. Re-insert Snap-Valve bulb into swab tube.

3

3. **Activate the device.** Bend bulb, snapping the Snap-Valve pin.

4

4. Lift the bulb up (about 1 – 2 inches) and squeeze the bulb to release the liquid into tube. Release pressure from the bulb (the bulb is like a dropper bulb) and replace bulb into the tube. Most liquid should be in the bottom of the tube.

5

5. Shake the tube gently to mix sample in the liquid.

6

6. Incubate at 30° ± 2°C for 7 hours. This is the **Enriched Sample**. Proceed to Step 2.

Step 2: Detection / Measurement

1

1. Allow **MicroSnap TOTAL Detection Device, MS-TOTAL100** to equilibrate to room temperature. Shake tube by tapping on the palm of your hand 5 times to bring liquid in the tube to the bottom of the tube.

2

2. Aseptically transfer 0.1ml (3 drops or to fill line) of **Enriched Sample** from **MicroSnap TOTAL Enrichment device (MS-TES100)** into **MicroSnap TOTAL Detection Device (MS-TOTAL100)**.

3

3. Activate **MicroSnap TOTAL Detection Device (MS-TOTAL100)** by breaking the Snap Valve pin with a snap and squeeze action. Squeeze the bulb to release the liquid into the tube. Liquid should be in the bottom of the tube.

4

4. **After activation**, shake the tube gently to mix sample in the liquid.

5

5. Insert **MicroSnap TOTAL Test (MS-TOTAL100)**, into a luminometer and initiate the measurement.

6

6. Record the results as RLUs and refer to Table 1 to interpret the results.



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