

# User Guide

## for microbial enumeration on surfaces with nomad Testers

### Introduction

The nomad Testers are simple, easy-to-use and ready-to-go devices for assessment of microbiological contamination levels when access to a laboratory, dedicated equipment or expert technique are limited.

Applications include microorganism enumeration in environmental waters, process waters, purified waters, equipment rinse waters, food and beverage products, manufacturing equipment including work surfaces for hygiene, environmental and/or process monitoring.

The nomad Swab Kit is a self-contained system, incorporating testers and swabs containing 18 ml of sterile, phosphate buffer into which are fitted swab assemblies.

The swab dilution buffer contain neutralising agents to counteract the adverse effect of any residual chlorine or quaternary ammonium compounds that may be present on surfaces after sanitation.

The test devices unique all-in-one design allows for sampling and testing, thus eliminating the need for a sterile transport container and an aseptic liquid transfer step.

The nomad Testers are based on the established microbiological culture method and membrane filtration technique. The Testers combine a 0.45  $\mu\text{m}$  membrane filter bonded to a culture media containing nutrient-pad, heat sealed into an easy-to-handle paddle.

The device configuration allows for the draw-through of 1 ml of sample to retain microorganisms on the filter surface, for subsequent culturing within its transparent plastic chamber. The filter is grid-marked to aid in counting the microbial colonies grown on its surface.

The system is designed for use in swab tests for measuring bacterial levels on flat or irregular surfaces. They can be used in a variety of applications in food production, food handling establishments, and other facilities to assess the efficiency of sanitation measures used to eradicate microbial contamination.

Isolated tests may provide useful information when the result is comparable to an accepted standard. Since facilities, processes and procedures differ from one another, regular testing that results in the build-up of historical data can be beneficial for assessing then preventing contamination risks by:

- Measuring the effectiveness of sanitary design, personnel practices, and operational methods.
- Providing information about indicator organisms, spoilage organisms, and/or pathogens of concern, so that appropriate corrective actions can be initiated to prevent potential microbial outbreaks.
- Acting as an early warning system for microbiological hazards in both the production and post-production environment
- Helping to identify harbourage niches and hot spots that may act as a source of contamination.
- Is a critical aspect of documenting the overall sanitary state of a facility.
- Validating the sanitation program and helps in determining the frequency required for cleaning and sanitation.

Consistency in the application of test protocols is important for building a solid data set.

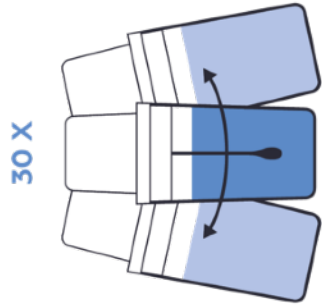
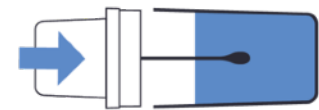
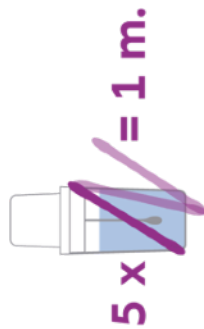
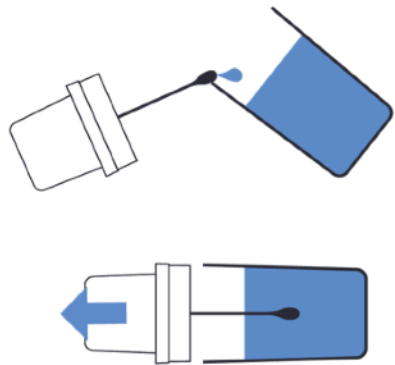
If a microbial monitoring program is in place, existing practices should supersede the recommendations provided in this guide.



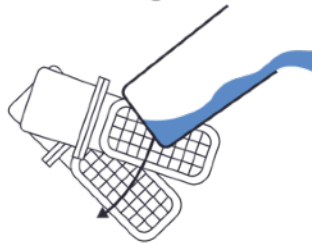
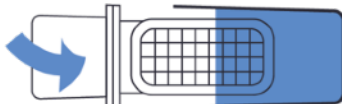
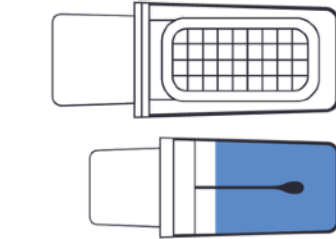
# Quick Start Guide - Surfaces / Lengths



## Sample



## Test



## Observe

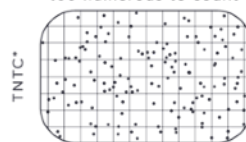
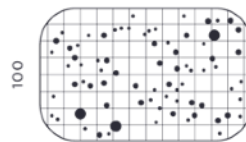
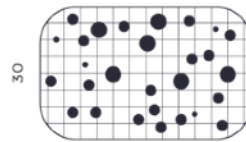
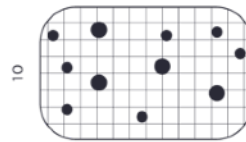
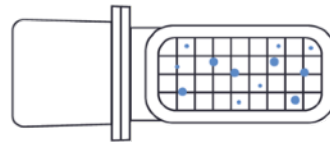


Coliforms

Total Count (HPC)

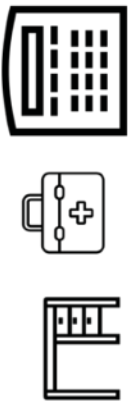
Yeast & Mold

35 °C / 95 °F	22-24 H
25-35 °C / 77-95 °F	48-72 H
28-32 °C / 82-90 °F	48-72 H

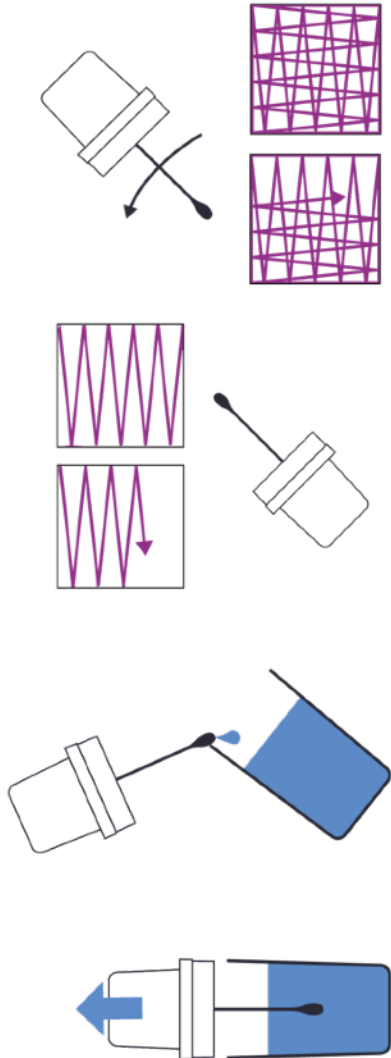




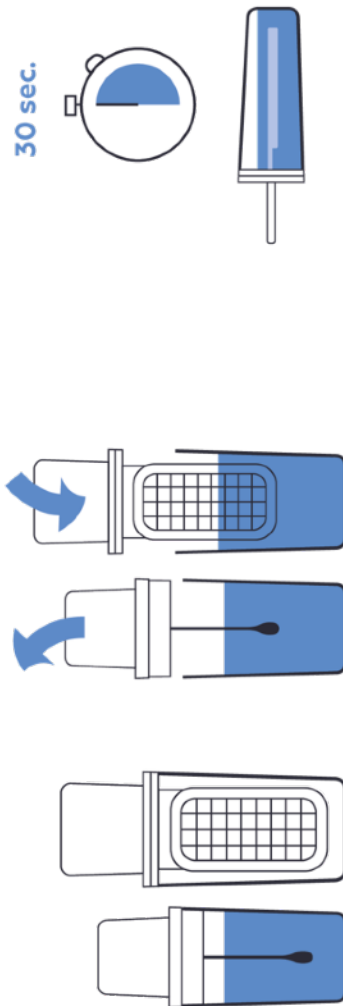
# Quick Start Guide - Surfaces / Areas



## Sample

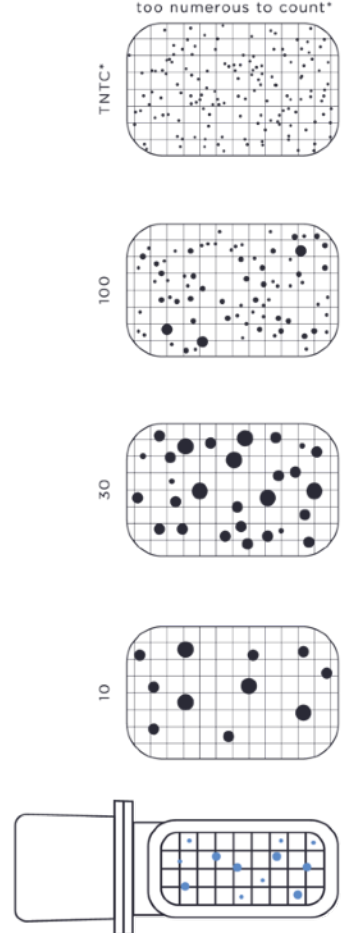


## Test



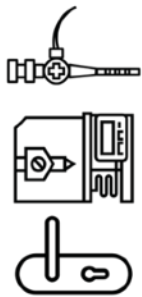
## Observe

	<b>Coliforms</b>	35 °C / 95 °F	22-24 H
	<b>Total Count (HPC)</b>	25-35 °C / 77-95 °F	48-72 H
	<b>Yeast &amp; Mold</b>	28-32 °C / 82-90 °F	48-72 H

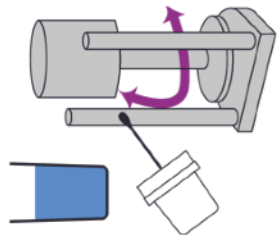
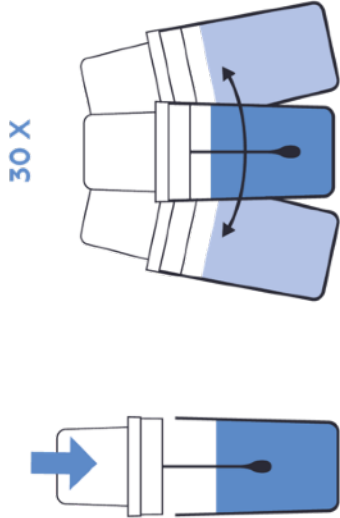
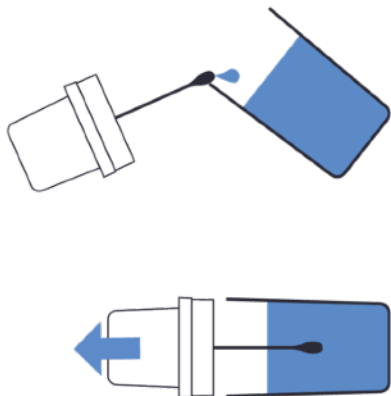




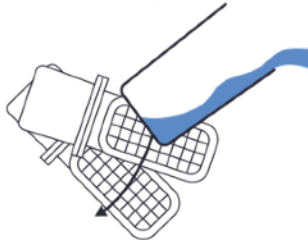
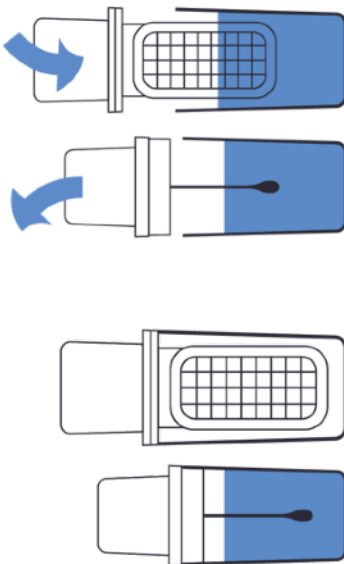
# Quick Start Guide - Surfaces / Objects



## Sample



## Test



## Observe

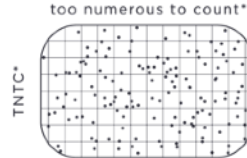
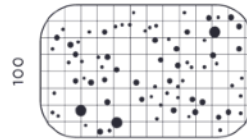
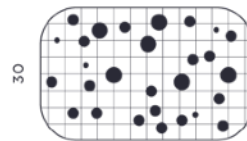
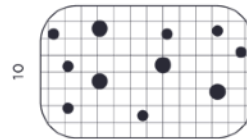
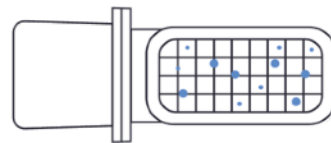


Coliforms

Total Count (HPC)

Yeast & Mold

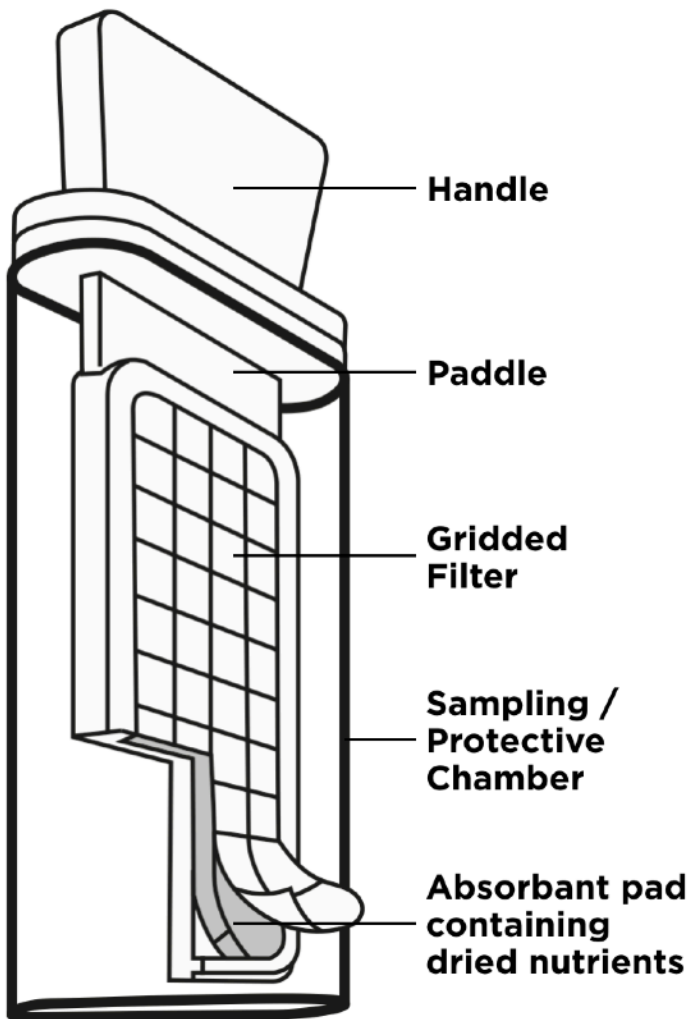
35 °C / 95 °F	22-24 H
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## Information Chapters

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## Tester components



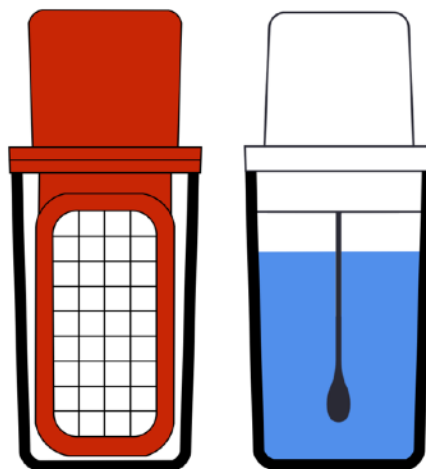
A Tester consists of two parts:

- An outer plastic chamber used to collect liquid samples and as a protective cover for the paddle.
- Each chamber carries 2 graduation marks, one for the 18 ml sample collection, another 1,8 ml mark facilitating 10 x dilutions
- A plastic dip test handle (paddle) with a gridded retentive filter and an absorbent pad.
- The pad contains the dehydrated nutrient medium for recovery of organisms.
- The 0.45  $\mu\text{m}$  membrane filter is bonded to the culture-media-containing nutrient-pad, heat sealed into an easy-to-handle paddle
- Each Tester assembly is packaged in a sealed plastic envelop.

A Swab consists of two parts:

- A plastic cap connected to a polyester swab,
- An outer chamber, identical to the tester chamber, containing pre-measured 18 ml of a sterile dilution and neutralising buffer solution.

A Swab Test Kit combines a Tester with a Swab.



# Instructions

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## Sampling

Collection of food, water and environmental samples may occur in a variety of locations.

Each location and reason for sampling will be associated with its own risks. It is important to make an assessment of these risks and put appropriate control measures in place before any sampling is carried out.

Examples of hazards include:

- Wet floors that present a slip hazard
- Working at heights when ladders/steps are required to reach sampling points
- Working in confined spaces when sampling from difficult-to-reach parts
- Exposure to aerosols when sampling from cooling towers and showers
- Exposure to irritant, corrosive or toxic chemicals
- Exposure to moving mechanical parts
- Lone working in isolated areas such as plant rooms

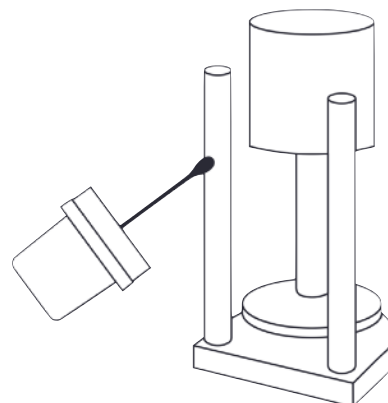
### A. Linear Sampling

Swabs are the preferred surface sampling method for rough, uneven, curved, hard-to-reach or damp surfaces such as surfaces of equipment, tools, cutting boards, keypads, inner tank surfaces, door handles.

They can also be used for flat surfaces on which the contamination level may vary from one part of the surface to another, for an indication of the average contamination level.

Linear sampling consists in applying the swab in a consistent way (swab angle with the surface, pressure applied to the swab, swab rotation during swabbing, sample length) along a chosen linear pattern.

Results should not be recorded as counts per surface area, since the swabbed surface is difficult to estimate, but may be recorded as « counts per Pattern-Name swab ».

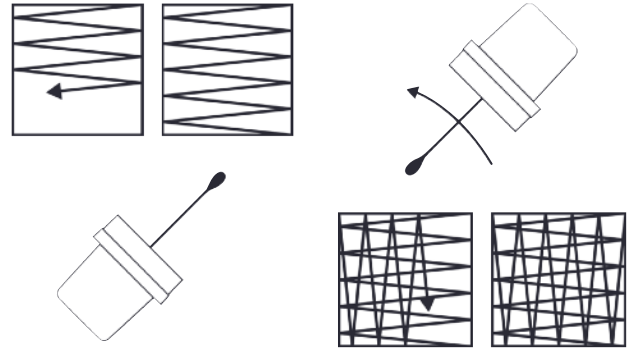


## B. Area Sampling

Swabs can be used on flat, dry and smooth surfaces, which are commonly tested with contact plates. The technique may then advantageously be put to use by swabbing multiple smaller surfaces than the 55 mm diameter disc (25 cm<sup>2</sup>) sampled by applying a contact plate.

A number of publications indicate that the 2 techniques, applied to flat, dry and smooth surfaces are not equivalent.

Results should be recorded as « counts per swabbed surface area » to avoid confusion between methods





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## Instructions for testing surfaces (Linear)

In testing the surfaces of equipment, cutting boards, etc., the nomad Swab Kit provides an overall picture of the test site (employing one swab with one tester only).

Sampling an isolated spot may not be representative of the whole test area.

The use of a swab provides high efficiency in penetrating crevices, rough and curved surfaces, and other hard-to-reach areas.

For sanitation monitoring, the application of the Swab/Tester combination therefore provides the method of choice for sampling these critical areas.

1. Write on the Swab case with indelible marker, the location of sampling.
2. Grasp the swab handle and, using an end-to-end rocking motion, remove swab from buffer case.
3. Roll tip of swab on inside of case to wring out excess buffer.
4. Select 5 areas of the surface to be monitored. Holding the swab firmly, draw a letter « N » on the surface, rotating the swab tip during procedure. Each straight section of the letter "N" drawn should be 6,5 cm ( 2 1/2") in length (the distance between 2 opposite corner of the swab chamber).
5. Repeat this test for the four remaining areas selected. Each letter "N" swabbed represents 20 cm (8 linear inches), thus 5 areas sampled is equal to 1 meter (40 linear inches) for each piece of equipment or surface area sampled. This method of sampling should be used each time the test is run in order to provide an equivalent comparison of results.  
Where the letter "N" cannot be applied (saw blades, slicers, etc.) swab 5 different areas of 20 cm (8 linear inches) each.
6. After swabbing the five selected points, insert swab firmly into case and shake 30 times to suspend the lifted organisms in the buffer.

Shortly after swabbing, within 1 hour if the sample is kept at ambient temperature, proceed with testing.

7. Open the Tester package, lift out the device.
8. Write on the Tester chamber with multi-surface marker the date, type and location of sampling.
9. Carefully remove the "paddle" from its case.  
To make it easier to remove the paddle, hold the Tester chamber with the membrane facing you and twist the handle towards you.  
Avoid touching the gridded filter surface at all times.
10. Remove and discard the swab.
11. Position the paddle over the buffer containing chamber and drop the paddle into the liquid.  
Quickly insert the paddle firmly into the chamber and lay the unit with membrane facing down onto a flat surface.  
Make sure the membrane is uniformly wetted, and while in this position, the unit should not be agitated.  
Allow 20 seconds for sample to be drawn through the filter and ensure there are no more bubbles coming out of the vent prior to removing the paddle from the sample.  
The filter should appear a uniform grey.

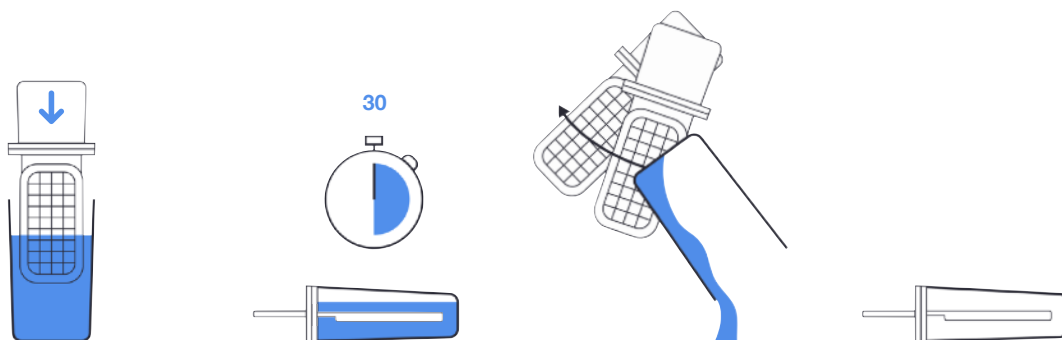
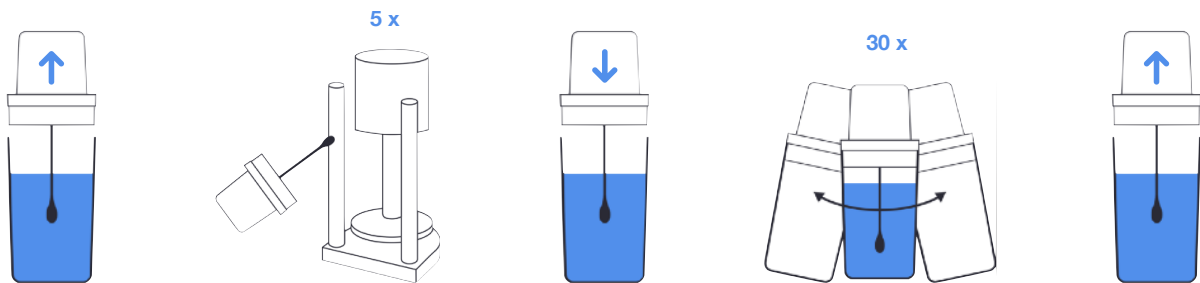
12. After 30 seconds have elapsed, remove paddle from case and shake once or twice vigorously to remove excess buffer.
13. Reinsert Tester paddle firmly into its dry chamber with the filter facing the opposite to the marking on the chamber. To prevent the paddle from drying out during incubation, it should be seated firmly in the case to form an air-tight seal.
14. Incubate the Tester, filter side down, using the time and temperature specified in the table shown in the "Culture-Incubation Guide".
15. For examination and counting please refer to the « Colony Counting Guide" section.

NOTE: Since only 1 ml from the 18 ml of buffer containing the organisms was drawn through the Tester filter, the realistic count for the area swabbed would be the count obtained X 18.

In principle, swabbing 1 linear meter with a 2,5 mm thickness corresponds to 25 cm<sup>2</sup> which is the surface sampled using a standard 55 mm contact plate.

However, the estimates of the surface actually swabbed can be inaccurate and the comparison with other techniques impractical.

The counts obtained, if this method is applied consistently are appropriate for many monitoring programs.



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## Instructions for testing surfaces (Area)

In testing flat surfaces of equipment, tanks, benches, the nomad Swab Kit provides an overall picture of the test surface (employing one swab with one tester only).

Sampling an isolated spot may not be representative of the whole test area.

The swab dilution buffer contain neutralising agents to counteract the adverse effect of any residual chlorine or quaternary ammonium compounds that may be present on surfaces after sanitation. For sanitation monitoring, the application of the Swab/Tester combination therefore provides a method of choice for sampling these critical areas.

1. Write on the Swab case with indelible marker, the location of sampling.
2. Grasp the swab handle and, using an end-to-end rocking motion, remove swab from buffer case.
3. Roll tip of swab on inside of case to wring out excess buffer.
4. Select 5 areas of the surface to be monitored.  
Holding the swab firmly, draw 5 « horizontal » zig-zags over a 2.5 cm (1 inch) square.  
Flip the swab over 180°.  
Turn the swab 90° and draw 5 « vertical » zig-zags over the same square area (the size of the swab handle)
5. Repeat this test for the four remaining areas selected. Each swabbed square represents approximately 6 cm<sup>2</sup> (1 inch square), thus 5 areas sampled is equal to approximately 30 cm<sup>2</sup> (4 1/2 square inches) of surface area sampled.  
The same method of sampling e.g. the number of zig-zags, should be used each time the test is run in order to provide an equivalent comparison of results.
6. After swabbing the five selected points, insert swab firmly into case and shake 30 times to suspend the lifted organisms in the buffer.

Shortly after swabbing, within 1 hour if the sample is kept at ambient temperature, proceed with testing.

7. Open the Tester package, lift out the device.
8. Write on the Tester chamber with multi-surface marker the date, type and location of sampling.
9. Carefully remove the "paddle" from its case.  
To make it easier to remove the paddle, hold the Tester chamber with the membrane facing you and twist the handle towards you.  
Avoid touching the gridded filter surface at all times.
10. Remove and discard the swab.
11. Position the paddle over the buffer containing chamber and drop the paddle into the liquid.  
Quickly insert the paddle firmly into the chamber and lay the unit with membrane facing down onto a flat surface.  
Make sure the membrane is uniformly wetted, and while in this position, the unit should not be agitated.  
Allow 20 seconds for sample to be drawn through the filter and ensure there are no more bubbles coming out of the vent prior to removing the paddle from the sample.  
The filter should appear a uniform grey.

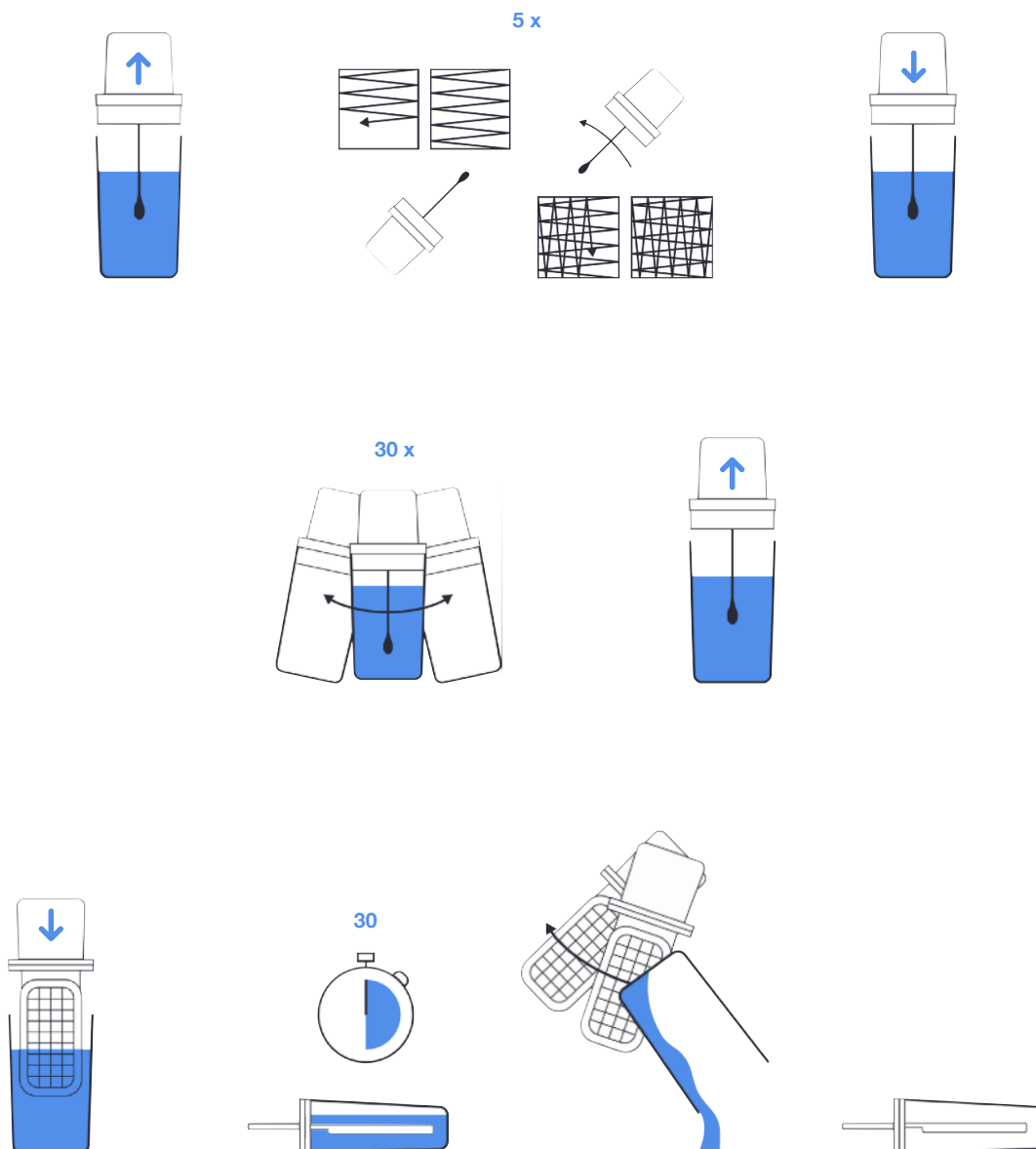
12. After 30 seconds have elapsed, remove paddle from case and shake once or twice vigorously to remove excess buffer.
13. Reinsert Tester paddle firmly into its dry chamber with the filter facing the opposite to the marking on the chamber. To prevent the paddle from drying out during incubation, it should be seated firmly in the case to form an air-tight seal.
14. Incubate the Tester, filter side down, using the time and temperature specified in the table shown in the "Culture-Incubation Guide".
15. For examination and counting please refer to the « Colony Counting Guide" section.

NOTE: Since only 1 ml from the 18 ml of buffer containing the organisms was drawn through the Tester filter, the realistic count for the area swabbed is the count obtained X 18.

In principle, 25 cm<sup>2</sup> of a surface should result in the same counts with the swab technique as with the contact plate technique.

However, a number of publications indicate that the 2 techniques are not equivalent.

Results should be recorded as « counts per swabbed surface area » to avoid confusion between methods



## Culture-Incubation Guide

When using the blue Tester i.e. performing coliform analysis, the time and temperature shown below must be followed.

The other Testers may be incubated at any temperature shown below as long as the temperature is consistently followed.

The mid-points of these ranges are preferable.

Time of incubation (except Coliforms) should be at least 48 hrs.

Incubation periods exceeding this will be dictated by practical time limitations.

The incubation period should be consistent.

Incubation		
	Temperature (°C / °F)	Duration (Hours)
<b>Blue (Coliforms)</b>	35 / 95	22-24
<b>Red (Total Count)</b>	25 / 77	72
or	35 / 95	48
Starved, heat or chemically stressed microorganisms	Room temperature	3-7 days
<b>Yellow (Yeast and Mould)</b>	28 / 82	72
or	32 / 90	48

## Colony Counting Guide

After incubation is complete, remove paddle from its chamber and examine the filter surface, preferably with an illuminated magnifier (5X to 10 X).

A Smartphone may also be used as a magnifier, in one of 2 ways:

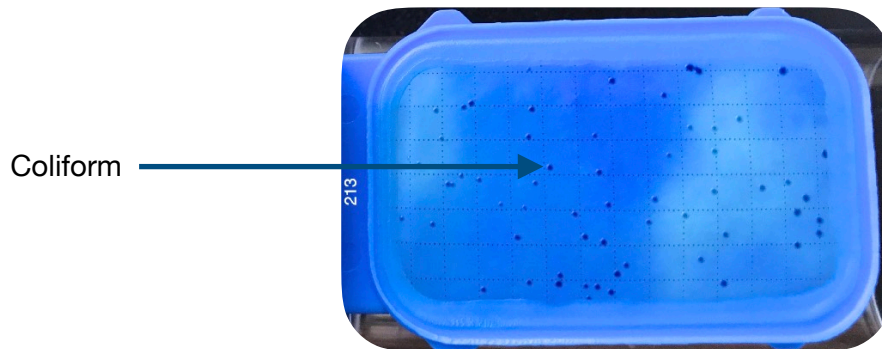
- Using the camera function with maximum zoom
- Taking a picture at a distance of 4 inches or 10 cm, then zooming in on the picture.  
This approach also allows the counting to be performed later or by another person.  
Make sure the date, type and location marked on the device are visible on the picture.

The appearance of the microbial colonies will vary, depending upon the type of device used and the organisms recovered.

Generally they will appear as follows:

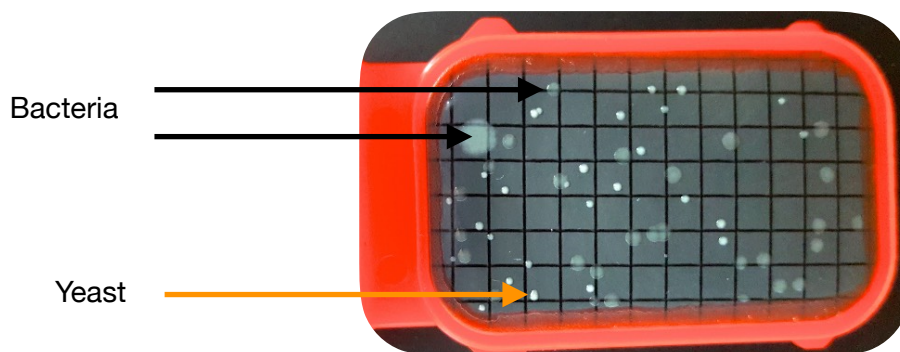
Blue nomad Testers:

- Coliform colonies are blue or blue/green.
- Non-coliform colonies may be green, grey, or cream.



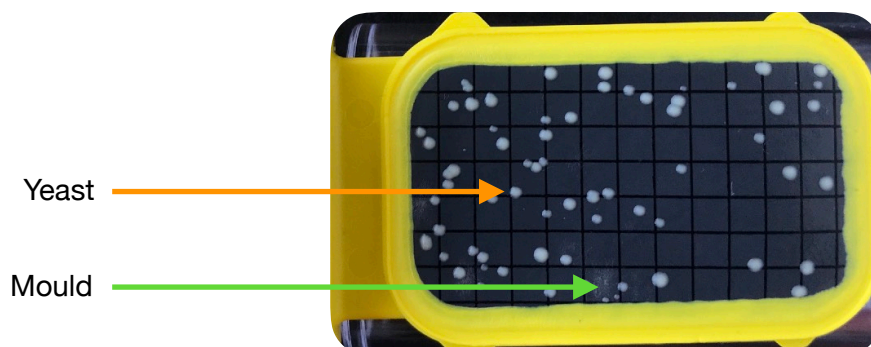
Red nomad Testers:

- Bacteria colonies appear glistening and translucent or transparent, circular or irregular in shape, with colours varying from colourless to white, cream yellow or occasionally pigmented.
- Mould colonies often appear filamentous whitish grey, with fuzzy edges. They usually turn into a different colour, from the center outwards
- Yeast colonies appear satiny, opaque, white coloured or may turn green over time.



Yellow nomad Testers:

- Mould colonies appear white, green or brown/black and filamentous.
- Bacterial colonies may appear but are usually smaller and more glistening and transparent than the yeast colonies.



## Counting Colonies

Colonies growing on the filter surface are counted as individual organisms.

In recording your count with the blue Tester for coliforms, count only the blue colonies.

Coliform and faecal coliforms are always reported as the number per 100 mL sample.

Therefore in undiluted samples, count the number of blue colonies obtained and multiply this result by 100. If the sample is diluted, multiply the 100 ml count by the appropriate dilution factor.

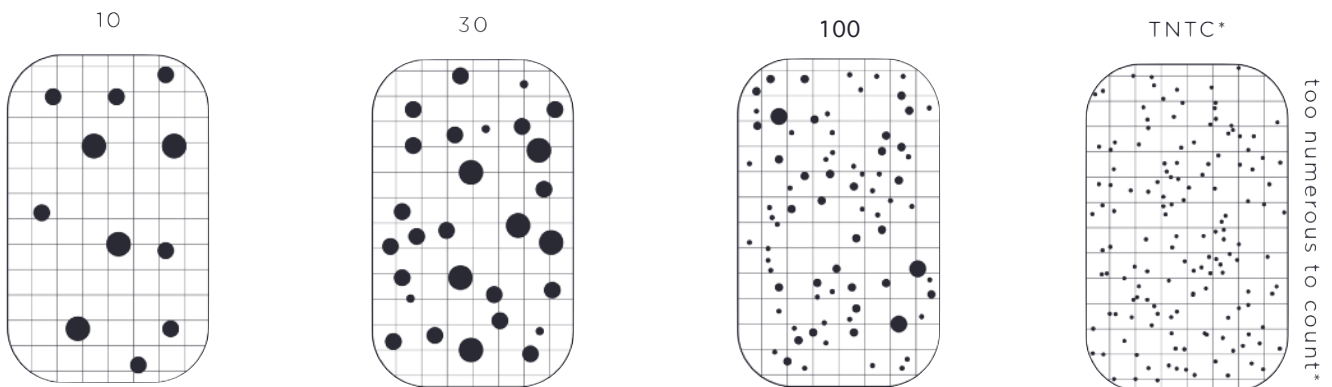
For all other samples, the count per ml is the generally accepted system for recording your results. Therefore, for non-diluted samples, the number of colonies observed on the filter will be the number recorded (as sample count/ml).

For diluted samples, the count obtained must be multiplied by the dilution factor.

For example:

- Number of colonies on filter = 60
- Sample dilution = 1:10 (dilution factor is 10)
- Sample count/mL =  $60 \times 10 = 600$  cfu/ml (colony forming units)

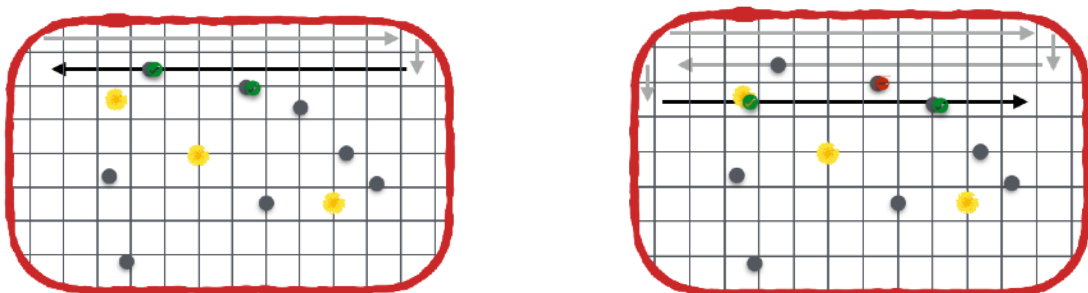
A rapid count estimate can be made by comparing the filter to the examples shown below.



### Precise counting technique

Starting from a corner of the grid-marked filter, follow the long grids and count.

When a colony has grown over a grid, to avoid double counts only count the colony the first time i.e. on the first pass.



Alternatively, starting from a corner of the grid-marked filter, follow the long grids and using a multi-surface pen, mark each colony position by a dot on the chamber. Once all the filter surface has been observed, count the dots.

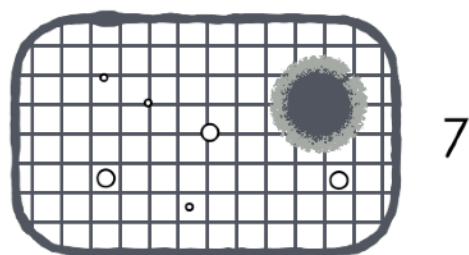
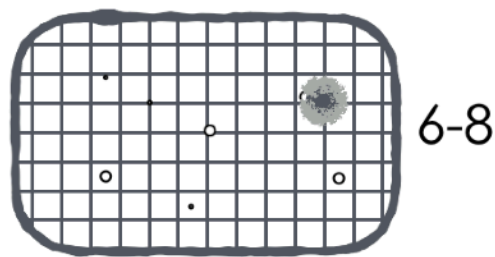
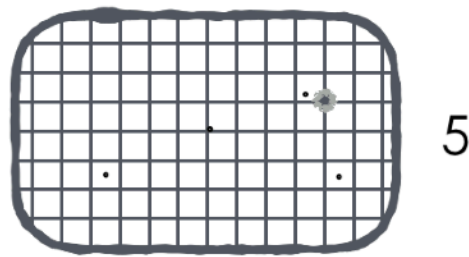
### Multiple readings

Some populations of microorganisms grow faster than others.

To improve the quality of the result, it is desirable to perform additional readings at incubation periods corresponding to  $2/3$  and  $4/3$  of the recommended incubation time.

These additional readings allow:

- to improve the counting of slow-growing populations whose colonies are difficult to see and count after a normal incubation period
- to reduce equivocal counts resulting from the fact that some fast-growing colonies overlap, join or come to confluence with others.



In this illustration, the observations are done on a Total Count Tester incubated at room temperature.

After the recommended incubation period, a mould has grown quickly and potentially overgrown a nearby bacteria colony.

In addition a small colony has appeared, located closely to a grid mark, which could be overlooked.

The final count is 8, while 8 individual colonies are not easy to observe on any of the individual readings.

In this case, the use of a multi-surface marker for marking each colony with a dot is recommended.



## Ordering Information

### **nomad Testers** for testing of liquids

Description	Color	Qty/Pk	Catalog #
HPC Total Count Tester	Red	25	NTRD TTC 25
Coliform Tester	Blue	25	NTRD COL 25
Yeast & Mold Tester	Yellow	25	NTRD YM0 25

### **nomad Swab Kits** for testing of surfaces

Description	Color	Qty/Pk	Catalog #
HPC Total Count Tester with Swab in Neutralising Buffer	Red	25	NTRK TTC 25
Coliform Tester with Swab in Neutralising Buffer	Blue	25	NTRK COL 25
Yeast & Mold Tester with Swab in Neutralising Buffer	Yellow	25	NTRK YM0 25
Swab in Neutralising Buffer	Red	25	NTRD SWB 25

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