Wagtech™

Potakit™ +

Basic Portable Water Quality Laboratory

ZI PTW 10030

Palintest
A Halma company

Who We Are

Over the last 20 years the **Wagtech™** name has become synonymous with water testing in the most extreme circumstances and remote locations.

Developed for a range of applications, from long term surveillance to rapid response testing in an emergency, the **Wagtech™** kits provide a robust solution to testing key water quality parameters in the field.

Acquired by **Palintest™** in 2011, the manufacture and support of the **Wagtech™** portable water quality laboratory range has now been integrated into the **Palintest™** product family.

Further information regarding the **Wagtech™** product range can be found at: **www.palintest.com**

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Potakit™ + Water Test Kit

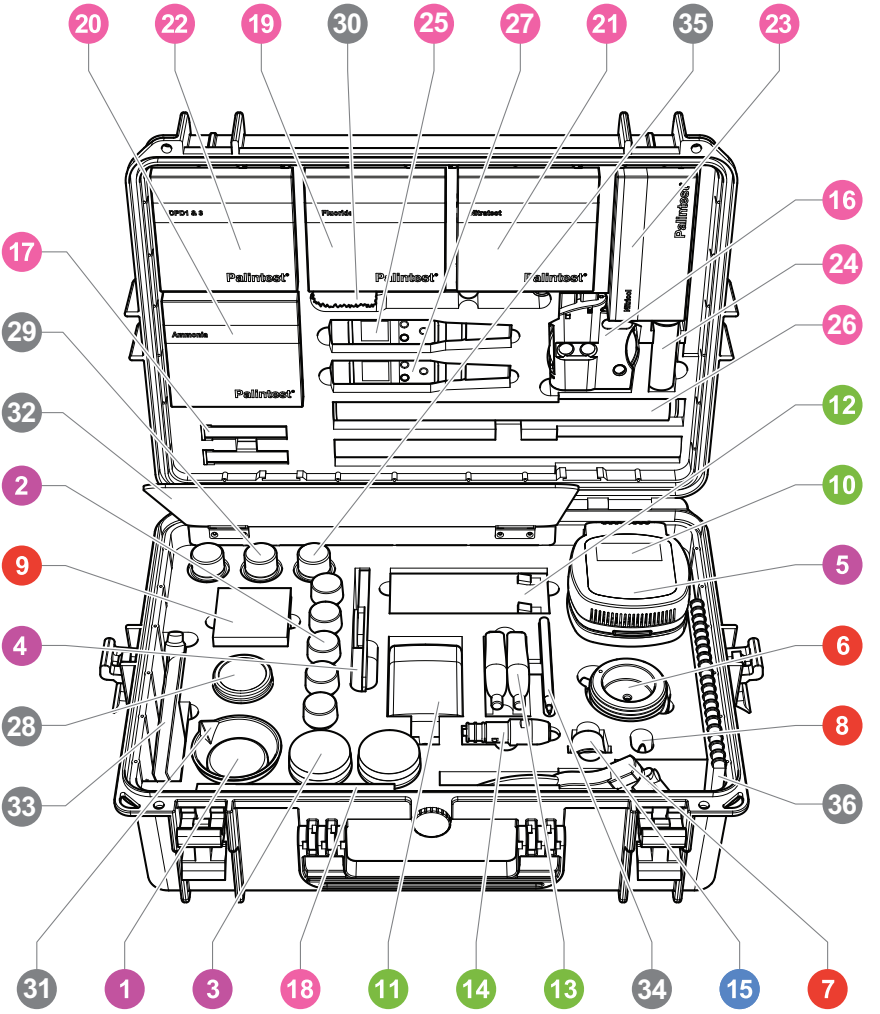


Fig 1. Potakit™ + Water test kit opened to show contents.
Coloured circles indicate the chapter colour in which their use is explained.

Function	Equipment
Preparing the Media/Petri Dishes	(1) **Membrane Lauryl Sulphate Broth (MLSB)** (2) **Media Measuring Device (MMD x 5)** (3) **Absorbent Pads x 2** (4) **Pad Dispenser** (5) **Petri Dishes (in incubator)**
Membrane Filtration	(6) **Membrane Filtration Unit** (7) **Hand Vacuum Pump** (8) **Forceps** (9) **47mm Membrane Filters**
Incubation and Incubator Operation	(10) **Incubator** (11) **Mains Power Supply** (12) **Battery** (13) **Battery Leads** (14) **Car Charging Lead**
Microbiological Results	(15) **Hand Lens**
Chlorine, Fluoride, Ammonia, Nitrate & Nitrite Testing	(16) **Contour Comparator** (17) **Comparator Cuvettes** (18) **Contour Comparator Discs** (19) **Reagent Tablets (Fluoride)** (20) **Reagent Tablets (Ammonia)** (21) **Reagent Tablets (Nitrate) 'Nitratest'** (22) **Reagent Tablets (Chlorine DPD) 'DPD1 & 3'** (23) **Reagent Tablets (Nitrite) 'Nitricol'** (24) **Sample Tube, 10/20mL**
pH Testing	(25) **Pocket pH Sensor**
Turbidity Testing	(26) **Double Length Turbidity Tubes**
Conductivity Testing	(27) **Pocket Conductivity Sensor**
Other Items	(28) **Dilution Tube - Sample Bottle Inside** (29) **Buffer Solution** (30) **Bottle Brush** (31) **Measuring Beaker** (32) **Work Surface** (33) **De-ion Pack** (34) **Pen** (35) **Conductivity solution** (36) **Instruction Manual**

2.0 Introduction

The **Wagtech Potakit™ +** is a portable water quality test kit. It has been designed primarily to test the microbiological quality of drinking water; assessing whether or not there has been faecal contamination of a water source. It allows the end user to test directly for Total and Faecal Coliforms.

The **Wagtech Potakit™ +** conforms to advice given by the **World Health Organisation (WHO)** for the field based testing of microbiological water quality. The parameters measured for, and techniques/ procedures used are based on accepted laboratory methods and are adapted for use in demanding field conditions.

More information on the **"WHO Guidelines for Drinking Water Quality"** can be found at **www.palintest.com**

As with all the kits in the Wagtech range, ease of use is integral to the design. The **Wagtech Potakit™ +** is suitable for use by technicians of all skill levels and this manual provides the essential information required to conduct rapid water quality testing in the field.

This instruction manual is also available in French, Spanish and Mandarin.

Additional advice and training is available upon request. Contact us directly at **support@palintest.com** or via your local representative.

2.1 Before You Use Your Kit

2.1.1 Microbiological Analysis of Drinking Water

Drinking water contaminated by faecal matter may contain pathogenic (disease causing) organisms and represent a risk to public health.

It is impractical to attempt to isolate specific pathogens because they are present in relatively small numbers compared with other types of microorganisms. Moreover, there are many types of pathogen and each requires a unique microbiological isolation technique. The accepted approach is to analyse for indicator organisms that inhabit the gut in large numbers and are excreted in human/animal faeces. The presence of these indicator organisms in water is evidence of faecal contamination and therefore a risk that pathogens are present. If indicator organisms are present in large numbers, the contamination is considered to be recent and/or severe.

The group of indicator bacteria tested for with the **Potakit™ +** are called Coliforms; more specifically the focus is on the enumeration of Thermotolerant Coliforms (sometimes called Faecal Coliforms). These are bacteria that originate from faecal sources. However the **Potakit™ +** is also capable of testing for Total Coliforms by simply selecting a different incubation temperature.

Thermotolerant Coliforms or Faecal Coliforms are used in water microbiological testing to denote coliform organisms which grow at 44 or 44.5°C and ferment lactose to produce acid and gas.

In practice, some organisms with these characteristics may not be of faecal origin and the term Thermotolerant Coliforms is therefore more correct and is becoming more commonly used. Nevertheless the presence of Thermotolerant Coliforms nearly always indicates faecal contamination.

Usually, more than 95% of Thermotolerant Coliforms isolated from water are the gut organism Escherichia coli (E. coli), the presence of which is definitive proof of faecal contamination.

As a result, it is often unnecessary to undertake further testing to confirm the specific presence of E. coli.

Total Coliforms refers to a large group of Gram-negative, rod-shaped bacteria that share several characteristics. The group includes Thermotolerant Coliforms and bacteria of faecal origin, as well as some bacteria that may be isolated from environmental sources.

Thus the presence of Total Coliforms may or may not indicate faecal contamination. In extreme cases a high count for the Total Coliform group may be associated with a low, or even zero, count for Thermotolerant Coliforms. Such a result would not necessarily indicate the presence of faecal contamination. It may be caused by entry of soil or organic matter into the water or by conditions suitable for the growth of other types of coliform bacteria. Generally, Total Coliforms are grown in or on a medium containing lactose at a temperature of 35 or 37°C.

Carrying out microbiological analysis of this sort therefore presents certain risks as it is highly likely that you will be handling equipment and materials that are potentially contaminated with harmful pathogens. This is especially relevant for the filter membranes, absorbent pads and petri dishes that are used in the test. For these reasons, general hygiene and aseptic procedures are of paramount importance and extra care must be taken when working in the field.

2.1.2 Overview of the Procedure for the Microbiological Analysis of Drinking Water

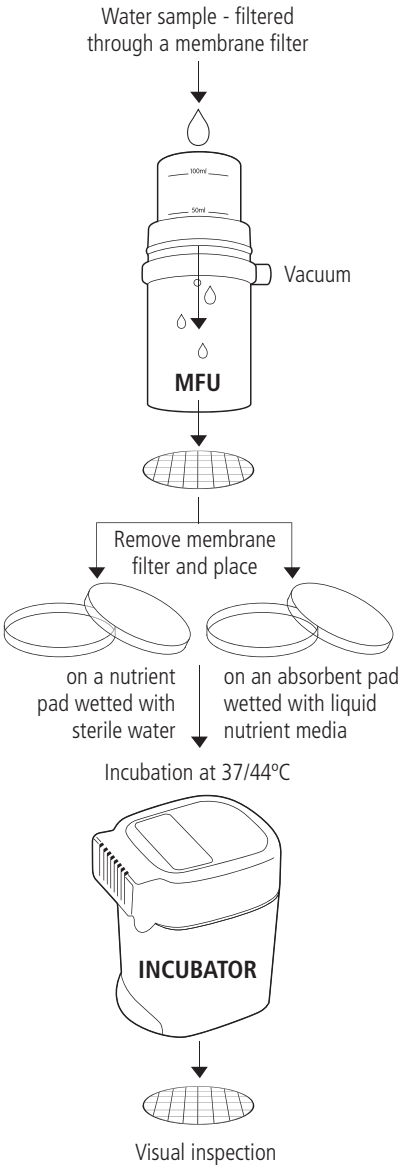
The Wagtech kits use classical laboratory techniques and equipment that have been adapted for use in the field. They conform fully with guidelines issued by **WHO** on accepted field-based methods for the microbiological analysis of drinking water.

The testing of water samples for coliform bacteria uses a method called **Membrane Filtration**.

In simple terms the process is as follows:

A known volume of sample water (100ml or less for highly contaminated samples) is filtered using a specific piece of apparatus called the **Membrane Filtration Unit (MFU)**.

Membrane Filtration Method



A vacuum hand pump attached to the MFU creates suction that pulls the sample water through a sterile membrane filter that sits in place in the MFU.

This membrane filter has small pores in it that allow the water to pass through easily but any bacteria present in the water are trapped on the surface of the filter membrane.

This filter is then removed and placed carefully onto an absorbent pad that sits in the base of a sterile petri dish.

The absorbent pad has been soaked in a liquid culture medium which provides nutrients for bacteria to grow whilst at the same time inhibiting growth of any non-target bacteria.

The petri dish is then placed in the portable incubator included in the **Potakit™ +** kits.

The temperature can be set to either 37 or 44°C, allowing the user to carry out tests for Total or Faecal (Thermotolerant) Coliforms.

The petri dishes are incubated for a minimum of 14 hours at optimum growth temperatures.

During this period the Coliform bacteria will multiply rapidly to form colonies that are visible to the naked eye.

Coliforms are identified by their ability to cause a colour change in the growth media when incubated. They will show up as yellow in colour against the red/pink background of the media.

The yellow colonies are counted and the results expressed as Colony Forming Units per 100ml of water - CFU/100ml (assuming sample size was 100ml).

Useful to Know

- Always wash your hands before carrying out your microbiological analysis, and again after handling potentially contaminated materials
- Never eat or drink while carrying out a microbiological test
- Never smoke while carrying out a microbiological test
- Do not directly touch any colonies in the petri dish
- Always hold the petri dishes by the sides and keep the lid on whenever possible
- Try to ensure that your workspace is clean and tidy - disinfect if possible (methanol)
- Ensure all open wounds are covered adequately
- Always sterilise the materials from the test before disposal and do not dispose of potentially contaminated materials directly into the environment

3.0 Preparation

Every effort should be made to keep the kit and all its components clean and free from contamination. At all times you should work in a manner that limits the chance of cross contamination of your samples.

3.1 Aseptic Procedure

There are specific techniques and methods that actively assist the user in trying to keep things clean and sterile. These are known as aseptic procedures or techniques.

This applies most importantly to the following items in the Wagtech test kit:

Membrane Filtration Unit:

The internal surface of the sample cup, the internal surface of the funnel and the filter base/bronze disc must be sterile before a microbiological test is carried out (section 3.2).

Petri Dishes:

The internal surfaces of the petri dishes will come into direct contact with the growth media during the microbiological test. They must be free from bacteria when the test begins. They can be sterilised in a variety of different ways (section 3.3).

Absorbent Pads:

The absorbent growth pads provide a platform for the liquid growth media and filter membrane which sit in the petri dish. There are specific instructions as to how they must be handled during the microbiological test (section 3.4).

Culture/Growth Media:

When preparing the media used for the test you must ensure that the water used to hydrate the powdered media is sterile. All the vessels used to prepare the media and into which the media is dispensed must also be sterilised in a specific manner (section 3.5).

Membrane Filters/Tweezers:

The membrane filters which capture the bacteria during the filtration process are supplied pre-sterilised. They must be handled using sterile tweezers and never with your fingers.

Useful to Know

- Methanol is highly flammable and is classed as 'Dangerous' for shipping purposes. For this reason it is not included as standard in the kit
- Methanol can be supplied separately from the kits but the freight and associated hazard charges may be expensive
- As an alcohol it can be challenging to find methanol in certain countries. Pharmacies, laboratories and hospitals are all possible local sources
- When methanol burns in the low oxygen conditions present in the sample cup of the MFU, a gas called formaldehyde is produced. This gas acts as a powerful disinfectant and ensures a complete sterilisation of the entire apparatus
- ONLY Methanol can be used to sterilise the MFU in the field. Ethanol or Methylated Spirits are not acceptable as they do not produce formaldehyde when ignited
- To ensure that the MFU is always ready for use it is a good idea to sterilise it after each analysis has been performed. The MFU must however be kept in sterile condition until the next time it is used

3.2 Sterilisation of the Membrane Filtration Unit

Upon receipt of your water test kit the **Membrane Filtration Unit (MFU)** must be washed thoroughly and then dried with a clean cloth or paper towel.

Prior to use, the MFU must also be sterilised. This is to reduce the risk of cross contamination of the water sample. It should be re-sterilised each time a new sample of water is analysed.

Sterilisation in the field can be difficult. A simple way of carrying out this sterilisation is with the use of **Methanol (Methyl Alcohol)**.

Items Required:

- Membrane Filtration Unit
- Methanol (Methyl Alcohol) - not supplied with kit
- Cigarette Lighter/Matches - not supplied with kit
- Paper Towels - not supplied with kit
- Plastic 1ml Pasteur Pipette

Procedure:

1 Using the Pasteur pipette add approximately 1ml of methanol into the stainless steel sampling cup.

2 Swirl the methanol around the inside of the sampling cup, coating as much of the internal surface as possible.

3 Holding the sampling cup facing away from you, use a lighter/match to ignite the methanol. The methanol will ignite instantly. Always exercise caution during this step. The methanol will burn with a pale blue flame; in bright sunlight it can be difficult to see this flame. However, the heat produced should confirm it has ignited.

4 Place the sampling cup base down on a flat surface while the methanol burns.

5 Assemble the filter funnel and silicone rubber base components of the MFU. Ensure that the filter funnel is inserted in the rubber base in the correct position for sterilisation (see below), which will leave a small gap between the bottom of the filter funnel and the silicone base.

Once the flame has virtually extinguished, invert the filter funnel and silicone rubber base components of the MFU and insert into sampling cup as shown below.

6 Under these conditions, formaldehyde gas is produced, which has excellent bactericidal properties. With the MFU in this position, formaldehyde gas can penetrate all contact areas, ensuring optimal coverage.

7 Leave for 15 minutes to ensure the formaldehyde gas carries out a complete sterilisation.

8 Remove filter funnel and base assembly from the sampling cup.

9 Pour any residual solution in the sampling cup away and re-insert the filter funnel and base assembly.

The Membrane Filtration Unit is now sterile and ready for use. It should be kept in the kit case until required.

IMPORTANT: This sterilisation procedure should be repeated each time a new water sample is to be analysed.

3.3 Sterilisation of the Aluminium Petri Dishes

Your Wagtech test kit is supplied as standard with a set of aluminium petri dishes. These are used during the microbiological analysis. They are used to hold an absorbent pad soaked in growth media.

A set of 20 petri dishes is supplied with each incubator complete with rack, which is used to lower them into the incubator and hold them in place during the incubation cycle.

The petri dishes are 50mm in diameter and are designed to accommodate the 47mm diameter absorbent pads and filter membranes used in the microbiological test.

They are manufactured from aluminium so that they can be re-sterilised and used again and again.

Before use it is important that these petri dishes are sterile. They can be sterilised in a number of different ways:

- Autoclave/Pressure Cooker @ 121ºC for 15 minutes
- Immersion in a pan of clean boiling water for 15 minutes then allow to air-dry
- Heating in conventional oven at a temperature of greater than 180ºC for 30 minutes

3.4 Preparation of the Aluminium Petri Dishes/Absorbent Pads

1 It is recommended that the absorbent pad dispenser is sterilised before use. Add a few drops of methanol to the contact area, wipe with a clean cloth and place on work surface to dry.

2 Attach the absorbent pad cartridge to the dispenser, locking it into place.

3 Remove the lid of the petri dish and dispense a pad into the dish by sliding back the grooved lever using your thumb.

4 Once the pad is safely in the petri dish, immediately replace the petri dish lid. Take care when handling the petri dish. Never touch the internal surfaces of the petri dish, always hold the dish by the side.

5 Repeat the procedure until the required number of petri dishes are prepared and store the dishes in the petri dish rack. Place the rack in the incubator for safe storage until ready for use in the field. The media is added to the absorbent pads when on site at the time of sample processing.

3.5 Media Preparation Procedure

The **Potakit™ +** is supplied with five **Media Measuring Devices (MMD)**. MMDs are pre-sterilised polypropylene containers. The blue screw cap lid features an integrated spoon/spatula.

The MMD is used to prepare the culture media in small quantities for optimum ease of use in the field.

Each MMD can hold 25ml of liquid media, enough for ten microbiological tests.

The MMD allows end-users to prepare sufficient media for daily requirements and removes the problems associated with preparing and storing large amounts of liquid media.

The MMDs can be re-sterilised after each use, without affecting product performance.

3.6 Preparing Culture Media in the Field using the Media Measuring Device (MMD)

Items Required:

- 38.1g Membrane Lauryl Sulphate Broth (MLSB)
- Media Measuring Device (MMD)
- 100ml distilled or clean water
- pH Meter
- Pressure Cooker/Steriliser/Autoclave
- Heat Source

1 If no distilled water is available, choose the cleanest water available e.g. rainwater, filtered water, or, if necessary, stand raw water in a container overnight. Do not use water which has been chlorinated. Boil the water for at least ten minutes, cover and allow it to cool down.

2 If using raw water then it may be necessary to filter 100ml of this water through the membrane filter using the membrane filtration unit (MFU) - see Section 7 MFU Operation. If the water is turbid, this step may need to be repeated. Prepare as much filtered water as you require.

3 Use the pH Meter to check the pH of water to be used to make up the liquid media. To make up MLSB the water should be pH 7.2 - 7.6. pH must be corrected or an alternative source of water will need to be found if the value is outside of the ideal range. Adjust the pH by using dilute sodium hydroxide solution (increases pH) or dilute hydrochloric acid (reduces pH).

4 Boil the distilled/clean water for 10 minutes and then allow it to cool.

5 Ensure that the MMD are sterile before beginning. MMDs can be sterilised upright using an autoclave. Screw on the lid so that it is secure but not completely air tight to prevent high pressure developing. Take care not to place the MMD directly onto the base of the autoclave. Sterilise at 121°C for 10 minutes. Alternatively, place MMD in a pressure cooker and maintain steam at pressure for 15 minutes.

6 Remove the MMD, allow to cool.

7 Once cool, unscrew the blue lid/spoon of the MMD. Take ten level spoonfuls of media from the 38.1g container of the MLSB and add to the MMD. Always hold via the lid and do not touch the spoon itself.

8 Fill the MMD with sterilised water to the lower lip and screw the lid on tightly.

9 Shake the MMD to aid dissolving of the powdered MLSB. Once dissolved a bright red/pink liquid will be produced.

10 Ideally, to minimise the risk of contamination, the MMD containing the liquid MLSB should now be sterilised. Undo the lid slightly so that it is secure but slightly loose and sterilise again as in Step 5.

11 Upon completion, remove the MMD and allow to cool. Tighten the tops securely and store in a cool, dark place until ready to use.

3.7 Tyndallisation of MMDs

In the event an autoclave or pressure cooker is not available then the sterilisation of the MMD containing the liquid MLSB media can be carried out using a pan of boiling water. This is a process called **Tyndallisation**.

When liquids are heated up to 100°C, the heat will kill the bacterial cells but the bacterial spores may survive.

Tyndallisation essentially consists of boiling the substance for 15 minutes for three days in a row. On the second day most of the spores that survived the first day will have germinated into bacterial cells. These cells will be killed by the second day's heating. The third day kills bacterial cells from late-germinating spores. During the waiting periods over the three days, the substance being sterilised is kept in warm, moist conditions that are conducive to germination of the spores. When the environment is conducive to the formation of cells from spores, the formation of spores from cells does not occur.

The Tyndallisation procedure can be summarised as follows:

- **DAY 1**: Place the MMD containing the MLSB into a pan or pot of boiling water, use a rack or stand if possible and try to prevent the MMD coming into contact with the base of the pan
- Boil for 15 minutes
- Leave the MLSB to cool and then stand at room temperature for 24 hours

- **DAY 2**: Once again heat the MLSB in a pan of boiling water for a further 15 minutes
- Leave the MLSB to cool and then stand at room temperature for 24 hours

- **DAY 3**: Repeat the immersion in boiling water for 15 minutes for a third and final time
- The MLSB media should now be sterile

3.8 Storage of Liquid Media

The sterile MLSB should be stable for up to 6 months if stored in a refrigerator. If no refrigerator is available the media can be stored for up to 3 months if kept in a dark place away from extremes of heat and moisture. However, if there are any signs of contamination e.g. yellowing, cloudiness etc., then it has become contaminated. It should be discarded and under no circumstances used in a microbiological test.

3.9 Pre-Prepared Media

It is possible to use pre-prepared growth media rather than prepare your own as described previously. The main advantages these options offer are that they save time and reduce the amount of equipment required. However it should be noted that they are generally more expensive in terms of cost per test, and their shelf-life is shorter. This makes shipping to, and then using in, remote locations more problematic. Some of the pre-prepared media also have specific storage requirements.

The most commonly used pre-prepared media options are:

Ampoules

Sterile **Ampoules** containing 2ml of dissolved media. Available in different varieties for the testing of a wide range of microorganisms. Simply unscrew the cap, pour the media onto the absorbent pad and discard the empty ampoule.

NutriDisks

For single use, a **NutriDisk** consists of a sterile plastic petri dish which includes an absorbent pad impregnated with dehydrated growth media. This is re-hydrated using sterile distilled water before use in the microbiological test.

The **NutriDisks** are available in different varieties for the testing of a wide range of microorganisms.

NutriDisks are the option most commonly used with the Wagtech Test kits. **NutriDisks** are larger than the standard aluminium petri dishes supplied in the kit but are still designed to fit into the standard petri dish rack. This allows up to 7 **NutriDisks** to be incubated at the same time.

To use the **NutriDisks** they are moistened with 3.0-3.5ml of sterile distilled water. They are then ready to use immediately. An excess ring of water surrounding the pad should be visible.

All **NutriDisk** types are supplied with the appropriate membrane filters, which are also pre-sterilised and individually packaged. The membrane filters are tailored to meet the special requirements of microbial detection and are available with 47mm or 50mm diameters.

See Appendix 4: Reagents & Consumables

Useful to Know

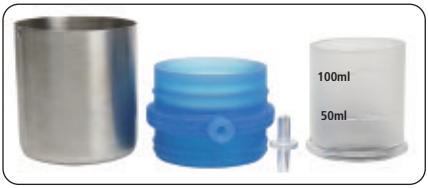
- Growth or culture media is a substance designed to support the growth of microorganisms (bacteria). The media is a vital part of the microbiological water quality test
- There are different types of media for growing different types of bacteria. The Wagtech kits use Membrane Lauryl Sulphate Broth (MLSB) as the growth media
- MLSB is a differential media which means it can distinguish one microorganism type from another growing on the same media. It uses the biochemical characteristics of a microorganism growing in the presence of specific nutrients or indicators (such as Phenol Red) added to the medium to visibly indicate the defining characteristics of a specific microorganism
- MLSB is the growth medium for Coliform bacteria and Escherichia coli (E.coli). It feeds coliform bacteria but inhibits the growth of any non-target organisms that may be present in the sample
- The MLSB in the kit is supplied as a fine powder. This increases its shelf life and makes transport and shipping easier. Preferably it should be stored in a dark environment away from extremes of heat and moisture
- Typically in its powder form the shelf life of the MLSB is 12 months
- In order to be used in the microbiological test the MLSB has to be prepared in liquid form. When water is added to powdered MLSB a deep red liquid is formed
- Coliforms are identified by their ability to cause a colour change in the growth media when incubated. MLSB contains lactose as the major carbon source, which during incubation is degraded to acid by E. coli and coliform bacteria; this is indicated by a change of the colony colour from red/pink to yellow

4.0 Taking A Sample

The optimum volume of sample is that which will allow the most accurate enumeration of bacteria. The technique of **membrane filtration** is unsuitable for natural waters containing very high levels of suspended material, sludges and sediments, all of which could block the filter before an adequate volume of water has been filtered.

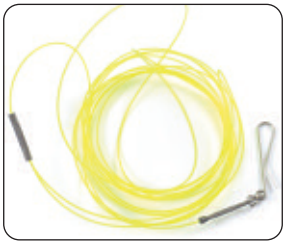
For potable or treated water samples the number of faecal coliform bacteria should ideally be zero in 100ml, indicating a microbiologically safe (or more accurately LOW RISK) water supply. The preferred sample volume is 100ml.

For raw source waters and partially treated waters, including those derived from ground water, it is sometimes useful to reduce the sample volume to obtain faecal coliform counts in the optimum range. This may be a reduction of the sample size to 50ml, or even 10ml in more contaminated water sources. To aid this, the filter funnel of the membrane filtration unit has two internal graduations at 50ml and 100ml.



4.1 Collecting the Sample for Analysis

Samples can be collected using the sterilised sampling cup of the **Membrane Filtration Unit (MFU)**. It is supplied with a cord to allow the sampling cup to be lowered into a water course, well or storage tank.



IMPORTANT: Always rinse the sterile sampling cup with some of your sample water before taking the final sample. This eliminates any residual methanol left over from the sterilisation process.

Care must be taken not to introduce floating matter or material from the edge of the water course into the water sample. It may be preferable to attach the sampling cable to the sterilised sampling cup and take the sample from a bridge or other overhanging location. Alternatively, the cup may be cast into the water from the edge and pulled slowly and carefully back towards the operator.

Alternatively, any suitable sterile container or sample bottle can be used in place of the sampling cup.

When sampling from a river or stream take the sample as near as possible to the main flow and not too close to the edge where the water may be still and unrepresentative of the sample as a whole.

When sampling from a tap or outlet that provides water for a consumer remove any tap attachment. Clean the tap/outlet with a dry cloth before allowing to run for 1 minute prior to sample collection.

Once collected, the sample must be processed immediately or as quickly as possible. The use of a portable field test kit makes this possible. However, if the delay between sample collection and analysis is between 2 and 6 hours, chill the sample rapidly to about 4°C with ice blocks in an insulated container/cool bag. Resuscitate the sample prior to full incubation using the automated routine contained within the Wagtech incubator software.

Even if the sample is kept cold, the maximum sample storage time is 6 hours. Analysis of samples not stored under these conditions or processed after a period of 6 hours are unlikely to reflect the bacteriological conditions at the time of sampling.

If chlorinated water samples are being collected, sodium thiosulphate (not supplied with kit) should be added to the sample bottles to neutralise chlorine.

5.0 Membrane Filtration of the Sample

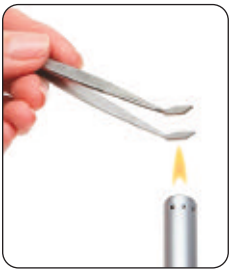
Items Required:

- Membrane Filtration Unit
- Pistol Grip Hand Vacuum Pump
- Forceps/Tweezers
- 47mm Membrane Filters (0.45µm pore size)
- MMD containing Liquid MLSB Media
- Petri Dishes containing Absorbent Pad
- Methanol (not supplied)
- Lighter/Matches (not supplied)

Ensure that at all times you work on the fold down work-surface that forms an integral part of the kit. Keep this area as clean as possible and before starting, wipe it down with a paper towel and a few drops of methanol.

1 Loosen the filter funnel and remove from the rubber base support. Invert the filter funnel and place it down on the clean work-surface. Also place the blue rubber base support down on the work-surface. Ensure that these items are only ever placed onto the clean work-surface.

2 Sterilise the forceps by passing them from side to side through a flame from a lighter and allow to cool. Take care not to heat for too long as this will cause sooty deposits to form.



3 Remove a sterile, individually wrapped membrane filter.

4 Peel back the transparent outer wrapper and use the sterile forceps to separate the white, gridded membrane filter from its blue backing paper and remove from the outer wrapper. Only ever grip the membrane filters at their edge.

5 Place the membrane filter directly onto the bronze filter support disc housed in the blue rubber base; ensure the gridded side is face-up. If the membrane tears or becomes contaminated, discard and use a fresh one.

6 Lock the membrane filter in place by pushing the filter funnel firmly into position in the blue rubber base. Take care to not touch the internal surface of the filter funnel with your hand.

7 Ensure the filter funnel is aligned correctly in the **'Filtration'** position, indicated by the graphic on the side of the filter funnel.

8 Pour the water sample into the filter funnel up to the 100ml graduation (or less if using a smaller sample).

9 Discard the excess water from the sampling cup then insert the filter funnel/base assembly into position in the sampling cup. Care is needed to prevent sample spillage from the filter funnel when assembling.

NOTE: If the sample is not to be collected in the sampling cup part of the membrane filtration unit, then the filter funnel/base assembly can be inserted into position in the sampling cup straight away.

10 Connect the hand vacuum pump and silicone tubing to the MFU.

11 Use the pump to create a vacuum and commence the filtration. The sample level in the filter funnel will fall rapidly.

Do not pump too many times so as to avoid drawing excess air through the membrane filter. The water passes through the pores in the membrane filter and is collected in the sampling cup. Any bacteria in the sample are collected on the surface of the membrane filter.

12 When all of the sample has been filtered, detach the vacuum pump and remove the filter funnel from the rubber base. The membrane filter is now ready to be removed and placed in the petri dish containing the absorbent pad and growth media.

13 Remove one of the sterile petri dishes previously prepared from the rack and place onto the work-surface. (Prepared petri dishes should contain an absorbent pad, if not, follow the procedure in Section 3.3).

14 Taking care to only handle the petri dish by the sides, remove the lid and place it on the work surface.

15 Take an MMD containing liquid MLSB media. Shake well then remove the blue screw lid and place it, lid-down, onto the work-surface.

16 Lift the petri dish containing the absorbent pad and hold between thumb and forefinger.

17 Take the MMD containing the liquid MLSB and carefully pour the media onto the absorbent pad in the petri dish in a single, decisive pouring motion. Always ensure that the petri dish is raised from the surface when pouring the media. Never use the plastic Pasteur pipettes to dispense media onto the pads.

18 Ensure the pad is well saturated with a small excess of MLSB visible at the edges. This should equate to between 2.5 and 3.0ml. If too much media is dispensed, simply pour the excess away. Place the petri dish back down on the work surface.

19 Use the sterile forceps to remove the membrane filter from the filtration unit.

20 Starting at the far edge of the petri dish, use a rolling motion to place the membrane filter on top of the absorbent pad. This will prevent air being trapped between the pad and membrane filter.

21 Replace the petri dish lid and label with sample number, place, date, time, etc. to identify in the rack.

22 Place the petri dish into the petri dish rack and repeat the process for all samples. Place the filled rack into the incubator for safe storage ready to start the incubation.

Ensure that the incubator is in the upright position at all times to avoid leakage of nutrient broth from petri dishes, paying particular attention when transporting.

5.1 Resuscitation Period

It is important to note that when the last sample has been processed, a resuscitation period of between one and four hours must be observed before incubation commences.

Environmental exposure can cause coliforms to become physiologically stressed. The resuscitation period allows coliforms to recover before culturing.

With this in mind it is essential to plan testing throughout the day, particularly if visiting multiple sample sites. Try to conduct all sample processing within a three hour window. This ensures a maximum resuscitation period of four hours.

The resuscitation period is especially relevant for water samples where the environmental exposure is due to chlorination.

The incubator operating software includes the capacity to have an initial resuscitation period as part of the standard incubation cycle.

5.2 Incubation Time

Incubate the samples for 18 hours at the desired temperature. Two preset incubation profiles are available on the Wagtech Incubator:

- To test for Total Coliforms incubate at 37ºC for 18 hours
- To test for Thermotolerant Coliforms incubate at 44ºC for 18 hours

Useful to Know

- It is preferable to run the incubator in-situ in the case
- Ensure the incubator lid is closed
- If powering from the battery only, ensure the case lid is closed to minimise power consumption
- Do not place the kit directly on the floor during incubation
- Do not incubate outdoors during periods of cold temperature
- Ensure that the petri dish rack is full during incubation, using empty dishes, to allow even heat distribution
- After switching on the incubator and selecting the desired temperature, allow a few minutes for the set-point to be reached and for temperature to stabilise. The incubator will show 'warming up' for a period of 30 minutes

6.0 Operation of Incubator

The **Potakit™ +** incubator is a high performance field incubator designed to deliver reliable Total and Thermotolerant Coliform results in even the most extreme circumstances. Providing at least 5 cycles of incubation under standard conditions when battery powered, the incubator is simple to operate and provides performance data throughout the incubation cycle.

6.1 Incubator Power Supply

The incubator can be powered in a variety of ways:

- Mains electricity 100-240V AC, via the mains adapter/charger unit
- 12V DC rechargeable battery (included) - sealed lead acid
- An external battery (12V DC) e.g. via the vehicle cigarette lighter attachment

6.1.1 Using the Incubator via the Mains Adapter/Charger

- If mains electricity supply is available, this method of operation is recommended
- Connect the cable of the mains adapter to the incubator via the socket on the left hand side of the incubator lid

- When connected to the mains power supply the incubator will show **'Charging'** with the battery icon
- Whenever possible ensure that the 12V battery is also connected to the incubator to provide charging/trickle charging. Connect the red and black cable connectors to the correct terminals on the battery and plug the cable into the right hand side of the incubator lid. This is advisable in areas where the mains supply may not be reliable. In the event mains power fails, the battery automatically provides power, thus continuing the incubation cycle
- Turn on the incubator by pressing the **POWER** button briefly

6.1.2 Powering the Incubator via the 12V DC Rechargeable Battery Only

- The incubator can also be powered solely by the 12V DC battery and a fully charged battery can provide up to five full incubation cycles
- Connect the red and black cable connectors to the correct terminals on the battery and plug the other end into the right hand side of the incubator lid

6.1.3 Powering the Incubator via an External 12V DC Source/Battery

- The incubator can also be powered via external supplies such as a vehicle/motorcycle battery or via the cigarette lighter socket of a vehicle

- Suitable cables/connectors are supplied in the kit for this purpose

6.2 Recharging the 12V DC Battery

Note: For optimal recharging, switch off the incubator.

To recharge the incubator battery:

- Connect the cable of the mains adapter to the incubator via the socket on the left hand side of the incubator lid
- Connect the red and black cable connectors to the correct terminals on the battery and plug the other end into the right hand side of the incubator lid
- Turn on mains electrical supply
- Charge for at least 8 hours
- The LED on the mains adapter indicates the status of **'charging'**:

Yellow = charging
Green = complete/trickle charging

Useful to Know

- Ideally the incubator should be recharged after each use, although as previously mentioned it is capable of running for up to 5 complete incubation cycles before this becomes necessary
- If this is not possible then ensure to charge the battery fully after prolonged periods of field use and try to leave the battery charged up when not using the kit
- Take care never to allow the battery to discharge completely as this will shorten its working life

6.3 Setup and Operation of Incubator

6.3.1 Start-up and Mode Page

- Ensure the incubator is connected to a reliable power supply. To switch the incubator on, press the **POWER** button and release

- A red LED in the right hand corner of the incubator lid indicates that power is supplied and the incubator is on. The backlight will illuminate automatically on key press.

- The initial screen is the **Mode** page and offers four options. Navigation between the options is carried out using the **UP/DOWN** buttons.
- The four options are:

  Incubation - select this option to carry out an incubation.

  Voice Instructions - select this option to hear useful audio instructions for key steps. Used with the prompt cards included.

  Setup - view or set User ID, Test Protocol, Speaker Volume, Date Format, Date, Time, Language, Check Calibration, Software Version and Resuscitation Period.

  Data Log - select this option to view reports of the last five incubation cycles.

6.3.2 Setup Menu

- The **Setup** menu allows the user to set the incubator preferences and validate performance
- To enter the Setup menu use the **UP/DOWN** keys to scroll to Setup on the Mode page

Once highlighted press **'OK'**. The following screen is displayed.

- Press **'YES'**
- Use the **UP/DOWN** key to scroll between the items in the Setup menu

User ID

Select or edit the username using alphanumeric characters. There are 8 separate User ID profiles.

- To create a new User ID, highlight one of the available profiles and press **'OK'**
- Select **'Edit'** to adjust User ID or **'Delete'** to reset to default
- Use the **UP/DOWN** buttons to show/change characters

- When the correct character is shown, release the key. The cursor moves to the next character automatically, up to a maximum of 12 characters
- Complete the process by pressing **'DONE'** briefly. To modify an existing User ID, highlight the ID and press **'OK'** briefly
- Choose **'Edit'** and use the **UP/DOWN** keys to select the required alphanumeric characters

To remove a character press **'Back'** or briefly press **'Back'** to allow editing of the right hand character.

- Once complete, press **'DONE'** briefly to return to the Setup menu
- To delete an existing User ID, highlight the ID and press **'OK'** briefly
- Choose **'Delete'** and press **'OK'**
- To return to the Setup menu, press **'BACK'**

Tests

Select the incubation temperature to set required time. There are two preset options available on the **Potakit +** incubator:

37ºC for 18 hours
44ºC for 18 hours

Incubation time can be adjusted to the required period using the **UP/DOWN** keys.

Speaker

To Set the incubator speaker volume:

- Highlight **'Speaker'** and press **'OK'**
- Use the **UP/DOWN** keys to adjust the volume
- Press **'OK'** to confirm and return to the Setup menu

Set Date Format

To set the preferred date format:

- Highlight **'Set Date Format'** and press **'OK'**
- Select **DD/MM/YY** or **MM/DD/YY** as required
- When correct press **'OK'**

Set Date

To set the Date:

- Highlight **'Set Date'** and press **'OK'**
- Use the **UP/DOWN** keys to set the day. When correct press **'OK'**
- Use the **UP/DOWN** keys to set the month. When correct press **'OK'**
- Use the **UP/DOWN** keys to set the year. When correct press **'OK'**

Set Time

To set the Time:

- Highlight **'Set Time'** and press **'OK'**
- Use the **UP/DOWN** keys to set the hour (24 hour clock format). When correct press **'OK'**
- Use the **UP/DOWN** keys to set the minutes. When correct press **'OK'** to return to the Setup menu

Set Language

To set the Language:

- Highlight **'Set Language'** and press **'OK'**
- Use the **UP/DOWN** keys to highlight the required language: English, French, Spanish and Mandarin
- When correct press **'OK'**

Check Calibration

Validate the temperature calibration of the incubator for performance review. The temperature is controlled by two laser-trimmed thermistors which are supplied factory calibrated and designed for long term stability.

There is no natural drift in the calibration of the incubator over time and hence no need to field-adjust the calibration.

Using two identical thermistors located separately provides a dual validation approach to performance monitoring. It is possible but unlikely that either thermistor may be damaged in normal use but the possibility of both thermistors being compromised in an identical manner is almost zero.

The Check Calibration function provides a simple electronic check that both thermistors are reading the same temperature as each other within a specified tolerance. If the comparison is out of range the display will show **Error 107: Validation**.

- Highlight **'Check Calibration'** and press **'OK'**
- The incubator will display **'Calibration Verified'** upon completion
- Press **'Back'** to return to the Setup menu

In the unlikely event of failure, return the incubator to your local service representative for attention, quoting the serial number shown on the base.

Version

View the software version number for identification and potential upgrade.

- Highlight **'Version'** and press **'OK'**
- Note the software version displayed

Resuscitation Period

To improve microbiological analysis performance, stressed samples should be subjected to a period of acclimation prior to incubation.

To automatically include a resuscitation period highlight **'On'** and press **'OK'**.

Resuscitation periods from 1-4 hours are set from the Incubation menu when set to **'On'**.

If resuscitation is not required highlight **'Off'** and press **'OK'**.

6.3.3 Incubation Menu

The **'Incubation'** menu is accessed from the **'MODE'** screen, confirms the incubator settings and starts the incubation cycle.

- Highlight **'Incubation'** and press **'OK'**
- Use the **UP/DOWN** keys to select numeric characters for the Sample ID
- Select each of the digits in turn and press **'OK'** until all four have been correctly set
- When complete press **'OK'** to confirm
- Use the **UP/DOWN** keys to select the appropriate User ID and press **'OK'** to confirm. (The User ID is created in the Setup menu)
- Use the **UP/DOWN** keys to select the desired incubation test profile. Press **'OK'** to confirm
- If Resuscitation Period is selected, adjust the temperature required using the **UP/DOWN** keys and press **'OK'**. The recommended value is 30°C
- Adjust the Resuscitation Period to between 1-4 hours depending on requirements
- The remaining battery life is displayed prior to incubation
- Press **'OK'** to start the incubation cycle

6.3.4 Voice Instructions Menu

The **Potakit +** Incubators all feature a high-volume audio speaker to deliver audio prompts. These prompts are used in conjunction with the included **Prompt Cards**. Together the two provide clear and concise instruction to key stages of the test procedure.

There are five standard prompt cards with corresponding audio, both indexed by letter and number.

The Prompt Cards are grouped as follows:

A0-A6: Preparing Culture Medium

B0-B6: Sterilising the Membrane Filtration Unit

C0-C6: Petri Dish Preparation

D0-D8: Membrane Filtration of the Sample

E0-E6: Microbiological Results

Audible prompts are available as standard in English, French, Spanish and Mandarin.

The **'Voice Instructions'** menu is accessed from the **'MODE'** screen.

- Highlight **'Voice Instructions'** and press **'OK'**
- Use the **UP/DOWN** keys to highlight the required language. English, French, Spanish or Mandarin
- When correct press **'OK'**
- Prompts start automatically at card A0. To move to a different prompt group use the **UP/DOWN** key to select B, C, D or E
- Press **'Next'** to move to the next prompt in the sequence
- Press and hold **'<(I<)'** to return to the Mode menu

NOTE: adjust volume of voice prompts to suit environmental conditions.

6.4 Incubating Samples

The incubation of samples can begin once the correct resuscitation period has been observed (if required), the incubator settings have been confirmed and a reliable power source is available.

- Ensure the full rack of petri dishes containing your samples is placed correctly inside the incubator and the lid is closed securely
- Complete the rack with empty petri dishes if less than 20 samples are required for optimum thermal efficiency
- Highlight **'Incubation'** from the Mode screen and press **'OK'**
- Enter the **'Sample ID'** number and press **'OK'** once correct
- Select the correct **'USER ID'** and press **'OK'**
- Select the correct test profile. Select **'37°C for 18 hours'** to test for Total Coliforms. Select **'44°C for 18 hours'** to test for Thermotolerant (Faecal) Coliforms
- The incubator will display the current power status as a percentage of battery remaining to ensure sufficient capacity is available to carry out incubation
- If insufficient battery is available the warning **'Error 110: Battery Low'** is displayed. Accept the error if an alternative power supply can be provided during incubation, alternatively wait until power is available prior to starting the process
- The **'Start'** screen will be displayed. Press **'OK'** to start the incubation
- The screen will scroll between two displays. The first displays time remaining in hours and current temperature. The second screen displays Sample/User ID and incubator cycle status
- The incubator has a default warm-up period of 30 minutes at the start of the incubation process. During this phase the incubator will display **'warming up'**
- Once the incubation cycle is complete the incubator will automatically switch off thus preventing the samples being incubated for longer than specified
- Stopping incubation manually can be carried out at any time:
- During incubation cycle press **'Stop'**
- The incubator will check **'Are you sure you want to stop?'**
- Select **'Yes'** to stop, **'No'** to continue incubation

Voice prompts can be accessed during incubation to guide through the stages of the microbiological process.

6.5 Data Log

The incubator provides selective recall of the last five incubation cycles, stored in chronological order.

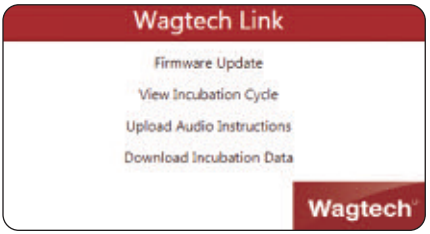
To review incubator data:

- Select **'Data Log'** from the Mode menu
- Use the **UP/DOWN** keys to select the required report and press **'OK'**
- The incubation report will show the following information:

16/04/14 (Start date for incubation)
21:00 (Start time for incubation)
37 degC - 18 Hr (Incubation Profile)
Wagtech (User ID)
2372 (Sample ID)

- To access the next incubation report press **'Next'**
- Press **'Back'** to return to the selection screen
- Press **'Back'** to return to the Mode screen

6.6 Wagtech Link Incubator App



The **Potakit +** incubator is provided with a micro-USB connection to allow Windows and Android devices (with internet access) to be connected for the following features:

- **Firmware Update:** Install the latest operating software simply to ensure maximum performance
- **View Incubation Cycle:** View data graphs for the last 100 incubation cycles identified by date, time and incubation profile
- **Upload Audio Instructions:** In addition to the standard language sets, additional audible prompts can be uploaded via the app for local language requirements
- **Download Incubation Data:** Download any stored incubation data in a Comma Separated Values (CSV) format for data manipulation and inclusion in reports

6.6.1 Firmware Update

The currently installed incubator software version is shown on start-up and can also be found in Setup>Version (see section 6.3.2). If the installed software requires update this can either be carried out via an approved service partner or by downloading the latest firmware from **www.palintest.com** and using the **Wagtech Link app**.

To update firmware:

- Connect the incubator to the device using the supplied USB cable
- Start the **Wagtech Link app**
- The device will connect automatically to the app and state **'Downloading Firmware'**
- Click the **'Firmware Update'** icon
- Select the **'Firmware Update'** and Click **'Send File'** to install
- The firmware file will upload with progress indicated by the progress bar
- Upon completion the message **'Upload Successful'** will appear and the incubator will automatically restart
- **'New software version'** will be displayed upon start-up

6.6.2 View Incubation Cycle

Up to 100 incubation cycles will be stored on the incubator in chronological order.

To view incubation data:

- Select **'View Incubation Data'**
- The dialogue screen will list the incubation cycles available for view listed by date/time, incubation protocol and number
- Choose the required cycle to view and press **'OK'**
- The app will plot incubator temperature on the top graph and power consumption on the lower graph over the entire incubator cycle
- Drag the progress icon to the right to see the trend throughout the cycle
- Click **'Fit to Window'** to see the entire incubation cycle on the device window at once

NOTE: only one incubation data file can be viewed; before opening additional cycles close the currently open view.

6.6.3 Upload Audio Instructions

The unique audible instructions included in the incubator, support new and experienced users alike in providing simple support for key steps in microbiological analysis.

Audible prompts are included in English, French, Spanish and Chinese (Mandarin). New prompts in local languages and dialects can be uploaded using the **Wagtech Link app** and **Audacity** software functionality.

To create correct format audio files for upload, download the freeware Audacity software **(www.audacity.sourceforge.net)** to your internet-enabled device.

The predefined format of acceptable audio files is:

File extension should be *.wav

Bit rate is 16bps

Sample rate frequency set to 8kHz

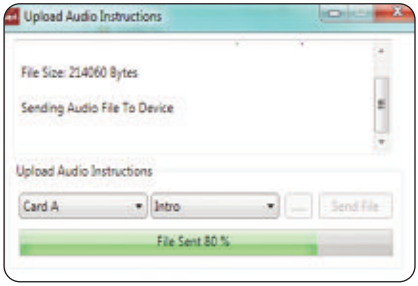
Max file size per clip is 640KB

Max length per clip is 40 seconds

Once audio files have been created for the individual prompts in the desired language/ dialect, save them to a memorable location.

To upload instructions via the **Wagtech Link app**:

- Connect the incubator via the USB cable
- Open the **Wagtech Link app**
- Select **'Upload Audio Instructions'**
- Select the **Prompt Card** required for the audible instructions (see section 6.3.4 and existing prompt cards)
- Select the address from Intro (0) to 6 for the audible prompt
- Select the file to upload to the specified location
- Select **'Upload'** and the progress bar will show the upload action



- Once updated, the selection of **'Custom'** will appear in the Voice Instructions menu and can be played in the usual way

6.6.4 Download Incubation Data

Water quality reports can be accented with the downloaded data from the incubator detailing the temperature profile and power consumption.

To download data in a **Comma Separated Values** (CSV) format:

- Connect the incubator via the **USB** cable
- Open the **Wagtech Link app**
- Select **'Download Incubation Data'**
- Choose **'All Data'** to download all available data or **'Selected Data'**
- Select the folder to store the downloaded data
- For **'Selected Data'** only, a list of available data sets will be shown. Multiple data sets can be selected using the shift or ctrl key
- Once all required data sets have been selected press **'Download'**
- **'Download progress'** will be shown on the dialogue box
- Upon completion the **'Download Finished'** message will appear

Downloaded **CSV files** can be opened with any spreadsheet or word processing package as a table of data for further manipulation.

6.7 Troubleshooting

The **Potakit +** incubator monitors performance during all stages of performance and provides information regarding unexpected condition as follows:

Error 102: Low Temp

The incubator has failed to reach the temperature set point within the specified time limit. Remove from areas of low temperature/high cooling and accept the error.

Error 103: High Temp

The incubator temperature has exceeded the set-point for a significant period. Remove from direct sunlight or high temperature conditions and accept the error.

Error 107: Validation

The internal validation of thermistor temperature monitoring has exceeded the tolerance of the device. Return to authorised service centre for attention as soon as possible.

Error 110: Battery Low

The 12V DC battery has insufficient battery capacity to complete the full incubation cycle. Prepare alternative power supply as soon as possible.

Error 111: Battery Critical

The 12V DC has approximately 10 minutes battery life remaining. Find alternative power source immediately.

Error 112: Power Loss

Unexpected power loss has interrupted incubation. Check for cause if possible and restart incubation when reliable power is available.

A full list of error codes is found in Appendix 2 - Troubleshooting.

Incubator Contamination

Should any hazardous material be spilt onto or into the incubator, cleaning and decontamination should only be carried out with a damp cloth and mild detergent.

Disconnect power during cleaning and do not invert the incubator.

Do not immerse the incubator under any circumstance.

Do not use acetone or any abrasive/aggressive/ hazardous cleaning agents. For advice regarding acceptable cleaning agents contact **support@palintest.com**

User Service

The **Potakit +** incubator contains no user serviceable parts. The automated calibration validation will confirm the incubator is operating correctly.

For service please contact your local Palintest representative or email to **support@palintest.com**

7.0 Microbiological Results

Bacteria in water are generally not present individually but as clumps or in association with particulate matter.

When enumerating bacteria in water it is not the number of individual bacteria present which are counted but the number of clumps of bacteria or the particles and their associated bacteria. Each clump or particle may have many bacteria associated with it.

Membrane filtration and colony count techniques assume that each bacterium, clump of bacteria, or particle with bacteria attached will give rise to a single visible colony. Each of these clumps or particles is therefore a **Colony Forming Unit (CFU)** and the results are expressed as colony forming units per unit volume. For standard volumes of sample this would be CFU/100ml.

This may vary depending on the type of water being tested and the volume of sample water actually filtered.

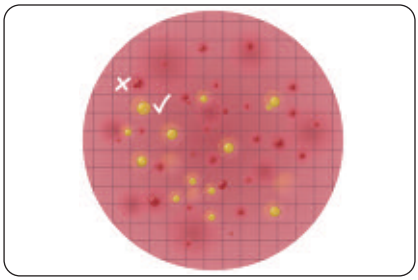
7.1 Enumeration Procedure

Following incubation, remove the petri dishes from the incubator. Take note of the temperature at which the samples were incubated.

Remove the petri dishes from the rack and place on the clean work-surface.

Remove the lids and, using the hand-lens if necessary, count all **yellow colonies**.

- If the incubation temperature was **37°C** then the yellow colonies represent a count for **Total Coliforms**
- If the incubation temperature was **44°C** then the yellow colonies represent a count for **Thermotolerant Coliforms**



7.2 Disposal of Used Materials

Before disposal, any materials used in the microbiological analysis must be made safe.

Potentially contaminated materials include Absorbent Pads and Filter Membranes.

Used pads and membranes cannot simply be thrown away after use as they represent a significant potential risk to public health.

Components can be made safe by sterilising the petri dishes and their contents. Ideally this should be carried out using an **autoclave** at 121°C for 15 minutes. Alternatively, a **pressure cooker** may be used. Once sterilised, the used pads and membranes can be incinerated. The petri dishes should be washed and re-sterilised, ready for the next use.

Useful to Know

- Count the colonies within a few minutes, as the colours are liable to change on cooling and standing
- Always try to have more than one person counting
- Try to count in the best available natural light - avoiding direct sunlight
- Count yellow colonies that are >1mm diameter
- DO NOT count colonies that are CLEAR, RED or ANY OTHER COLOUR - these bacteria do not ferment lactose and are not Thermotolerant Coliforms
- Gridded membrane filters permit easier counting when large numbers of colonies are visible
- Count colonies systematically, column by column in the grid
- Where there are too many colonies to count or it is difficult to see individual colonies clearly, mark the result as "Too Numerous To Count" (TNTC)

8.1 Jackson Turbidity Tubes

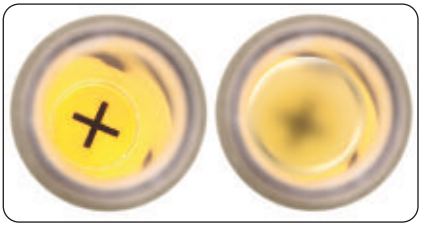
The two-part **Turbidity Tube** provides a simple and intuitive method of determining turbidity using a visual approach. Higher values of turbidity can be determined using the base section only, lower values require both sections.

8.1.1 Taking Turbidity Measurements

Connect the two parts of the Turbidity Tube together if the result is expected to be less than 20 **JTU**.

PIace the Turbidity Tube in a position where the base containing the cross can be observed.

Pour sample slowly into the Turbidity Tube until the cross is not clearly visible.



Read the turbidity value from the level of sample against the scale etched on the tube. Estimate the result if the level is between values.

Empty the Turbidity Tube and rinse with clean water prior to storage.

9.0 Pocket pH Sensor

The **Pocket pH Sensor** is a waterproof digital instrument featuring dual display of pH and temperature, plus automatic buffer recognition for simple calibration.

9.1 Taking pH Measurements

Remove the protective cap and press **ON/OFF** to switch the meter on.

Dip the electrode into 2-3cm of sample, rotate gently and wait for the reading to stabilise. The protective cap is a useful sample container.

Note the reading or press **HOLD/ENT** to store the reading on the display.

Press **HOLD/ENT** to return the display to normal mode.

After measurement rinse the electrode with clean water.

Store the electrode with a moist piece of paper in the protective cap if possible to avoid the electrode drying out.

9.1.1 Calibrating the pH Probe

Prepare the calibration solutions - A single point or up to three points may be used for calibration.

Remove the protective cap and press **ON/OFF** to switch the meter on.

Press **CAL** to enter calibration mode.

The upper display will show the actual reading, the lower display will show the buffer value.

Insert the probe into 2-3cm of buffer solution and stir/rotate gently. The protective cap can be used to hold the buffer.

Allow the reading to stabilise and press **HOLD/ENT** to confirm the value.

Rinse the electrode with clean water and repeat as required with additional buffers.

Press **CAL** to exit calibration mode at any point.

9.1.2 Calibrating Temperature

ON/OFF to switch the meter on.

Press **HOLD/ENT**

Press and hold **CAL** for 3 seconds to enter temperature calibration mode.

Press **CAL** to select **°C** or **°F** and the upper display will flash.

Insert the probe into a solution of known temperature and press **HOLD/ENT** to adjust the reading.

After 5 seconds the calibration will be stored and the meter will revert to normal mode.

9.1.3 Changing Batteries

Unscrew top casing to expose the battery compartment.

Replace with 4 **'A76'** button cell batteries, noting polarity.

Replace top casing.

9.1.4 Error Codes

- **Er.0** - temperature calibration is out of range. Use solution from 0-50°C
- **Er.1** - pH calibration solutions are out of range. Replace buffers with fresh. If problem persists replace pH electrode

10.0 Pocket Conductivity Sensor

The **Pocket Conductivity Sensor** is a waterproof digital instrument featuring dual display of Conductivity/Total Dissolved Solids (TDS) and Temperature.

10.1 Before You Begin

Remove the electrode's protective cap. Soak the electrode for a few minutes in alcohol to remove any oil stains on the electrodes. Rinse thoroughly with de-ionised water and shake to dry.

Press **ON/OFF** key to switch on the tester.

10.1.1 Range Selection

1 Switch off the tester. Press and hold **°C/°F** key and then switch on the tester using **ON/OFF** key. Release **°C/°F** key.

2 The LCD shows the currently selected Range (the default is **AUTO**) in the lower display. The upper display shows the maximum possible reading for the selected range. Press **HOLD** key repeatedly until you see the required range (PU, LO or HI).

10.1.2 Measurement

1 Press the **ON/OFF** key to switch on the tester. The **'MEAS'** indicator appears when the tester is in measurement mode.

2 Dip the electrode into the test solution making sure that it is fully immersed. Stir to clear any trapped air bubbles from the electrode and let the reading stabilise.

3 The upper display shows the main reading (conductivity/TDS) of the solution, automatically temperature compensated (ATC) to normalised temperature of 25°C. The lower display shows the temperature of the solution.

10.1.3 Calibration

Auto Calibration

1 Make sure the tester is in measuring mode. Press **INC** or **DEC** key to enter conductivity calibration mode.

2 **'CAL'** indicator appears in LCD. The display briefly shows **'CAL'** and the number of points the tester will be calibrated.

3 The upper display shows the conductivity reading and the lower display sequentially shows calibration standard values 84µS, 1413µS & 12.88mS if the measuring range of the tester is set to **AUTO**.

4 Rinse the electrode with the calibration standard that you intend to calibrate and then dip the electrode in the other beaker with same calibration standard. Swirl gently to create a homogenous sample and allow time for the reading to stabilise.

5 Press **HOLD/ENT** key to confirm the calibration. LCD shows **'CO'** for 2 seconds.

6 For multi-point calibration, the tester goes to the next calibration point. Rinse the electrode in de-ionised water and repeat steps 4 & 5 with next calibration standard solution.

Manual Calibration

1 Make sure the tester is in measuring mode. Rinse the electrode with the calibration standard that you intend to calibrate and then dip the electrode in the other beaker with same calibration standard. Swirl gently and allow time for the reading to stabilise.

2 Press **INC** or **DEC** key to enter calibration mode. The **'CAL'** indicator appears in LCD.

3 The upper display shows the measured conductivity/TDS reading of the solution based on previous calibration (if any) and the lower display shows the default (uncalibrated) conductivity/TDS reading. Note: The tester shows error message **'Er.1'**:

(a) If the reading is over range (Or) of selected measuring range of the tester, or

(b) If the default (uncalibrated) reading is not within the acceptable calibration standard range. Use **INC** and **DEC** keys to adjust the upper display to the correct conductivity/TDS value of the calibration solution.

4 Wait for 5 seconds for the tester to automatically confirm the calibration by displaying **'CO'** and return to the measurement mode.

10.1.4 TDS Factor Setting

The factory default TDS factor is 0.71. You can adjust the TDS factor to suit different samples of your applications.

To change TDS factor:

1 Make sure the tester is in measurement mode. Press **HOLD** key to bring the tester to the **HOLD** mode.

2 Press **INC** or **DEC** key to enter the TDS factor setting mode.

3 The upper & lower displays of LCD show the last configured TDS factor. The upper display is adjustable. Use the **INC** or **DEC** key to adjust the TDS factor. The adjustable range is 0.4 to 1.0.

4 Wait for 5 seconds for the tester to automatically confirm the new setting by displaying **'CO'** and return to the measurement mode.

10.1.5 Temperature Calibration

Temperature calibration need not be performed every time, unless the temperature reading differs from that of an accurate thermometer. If temperature calibration is performed, Conductivity/TDS calibration is mandatory.

1 Make sure the tester is in measuring mode. If required, press **°C/°F** key to select the desired unit of measurement for temperature (Celsius or Fahrenheit). Dip the tester into a solution of known temperature and allow time for the temperature reading to stabilise.

2 Press **INC** or **DEC** key to bring the tester to the calibration mode. **'CAL'** indicator appears in LCD. Immediately press **°C/°F** key to switch to the temperature calibration mode.

3 The upper display shows the measured temperature reading based on the last set offset (if any) and the lower display shows the default (uncalibrated) temperature reading based on factory settings. Use **INC** and **DEC** keys to adjust the upper temperature reading to the known temperature value of the solution.

4 Wait for 5 seconds for the tester to automatically confirm the temperature calibration value by displaying **'CO'** and return to the measurement mode.

10.1.6 Changing Batteries

Replace the batteries when the low battery indicator starts blinking.

1 Open the battery compartment lid (with attached lanyard loop).

2 Remove old batteries by pulling plastic ribbon. Replace with fresh ones.

11.0 Contour Comparator

The Contour Comparator provides visual testing for a wide range of parameters for drinking water testing in a compact and easy-to-use platform. Test methods are carried out using comparator grade tablet reagents for portability and stability in the field.

11.1.1 Free Chlorine

1 Select the Chlorine disk with the desired range and place in the Contour body.

2 Rinse tubes with sample leaving a few drops in the measurement tube.

3 Add one DPD 1 to the sample tube and crush to a paste.

4 Add sample to the tube to the 10ml mark and dissolve any remaining particles.

5 Place the tube in the measurement position.

6 Fill the second tube with sample and place in the blank position.

7 Facing a good light source, preferably northerly daylight, rotate the disk until the colours match and record the result from the lower right hand window immediately as mg/l Cl_2.

11.1.2 Total Chlorine

1 Add one DPD 3 tablet to the Free Chlorine sample and crush to dissolve.

2 Wait for 2 minutes to allow colour development.

3 Match the colours as before and record the result as mg/l Cl_2.

11.1.3 Combined Chlorine

Combined Chlorine =
Total Chlorine - Free Chlorine

11.1.4 High Chlorine Levels

1 If the colour disappears quickly after adding the sample the chlorine level is too high. Dilute the sample with chlorine free water until the colour persists.

11.2.1 Ammonia

1 Fill square test tube with sample to the 10ml mark.

2 Add one Ammonia No 1 tablet and one Ammonia No 2 tablet, crush and mix to dissolve.

3 Stand for 10 minutes to allow colour development.

4 Place the test tube in the Comparator and match against the disc in the usual manner (see Comparator instructions).

5 The disc reading represents the ammonia concentration present in the sample as milligrams per litre N.

11.3.1 Nitrite

1 Fill square test tube with sample to the 10ml mark.

2 Add one Nitricol Tablet, crush and mix to dissolve.

3 Stand for 10 minutes to allow full colour development.

4 Place the test tube in the Comparator and match against the disc in the usual manner (see Comparator instructions).

5 The disc reading represents the nitrite nitrogen concentration present in the sample as milligrams per litre N.

To convert Nitrite Nitrogen (N) to Nitrite ion (NO_2) multiply the result by 3.3.

11.4.1 Nitrate

1 Take a clean Nitratest tube and add 1ml of sample using the measuring syringe. Fill the Nitratest tube to the 20ml mark with de-ionised water.

2 Add one level spoonful of Nitratest Powder and one Nitratest Tablet. Do not crush the tablet. Replace screw cap and shake tube well for exactly one minute.

3 Allow tube to stand for about one minute then gently invert three or four times to aid flocculation. Allow tube to stand for at least two minutes to ensure complete settlement.

4 Remove screw cap and wipe around the top of the tube with a clean tissue. Carefully decant the clear solution into a square test tube, filling to the 10ml mark.

5 Add one Nitricol Tablet, crush and mix to dissolve.

6 Stand for 10 minutes to allow full colour development.

7 Place the test tube in the Comparator and match against the disc in the usual manner (see Comparator instructions).

8 The disc reading represents the nitrate nitrogen concentration present in the original sample as milligrams per litre N.

To convert Nitrate Nitrogen (N) to Nitrate (NO_3) multiply the result by 4.4.

11.5.1 Fluoride

1 Fill square test tube with sample to the 10ml mark.

2 Add one Fluoride No 1 tablet, crush and mix to dissolve.

3 Add one Fluoride No 2 tablet, crush and mix to dissolve.

4 Stand for five minutes to allow full colour development.

5 Place the test tube in the Comparator and match against the disc in the usual manner (see Comparator instructions).

6 The disc reading represents the fluoride concentration present in the sample as milligrams per litre F.

Measurement of arsenic is critical for health and well-being due to the chronic effects of arsenic-contaminated drinking water.

The Visual Arsenic Detection Kit (VCDK) is supplied in a separate case.

The kit contains all components for the analysis of arsenic in water, with sufficient reagents for 100 tests and a full instruction manual.

Note: Field testing requires careful planning

Ensure you have considered the following additional items that may be required in addition to the **Potakit™ +** contents:

- Sterilised sampling bottles/**Wagsac** sampling bags
- Result form/log book
- Methanol
- Non-electric autoclave/pressure cooker
- Cigarette lighter
- Paper towel
- Liquid detergent
- De-ionised water
- Waste disposal bags
- Cool box/icepacks

Before leaving for the field, ensure the following has been carried out:

- The equipment is sterilised wherever possible
- Sufficient petri dishes, absorbent pads, membrane filters, culture media and reagents are available
- Prepare MLSB media in Media Measuring Device (MMD) - enough for one day's testing. If using previously prepared media ensure the media is still bright pink and is not cloudy
- If time is available, prepare the petri dishes with pads before travelling to the field. This also allows absorbent pads to avoid contamination in the field
- If dishes are prepared before departure, an additional one or two are recommended for potential mishaps in the field
- Sterilise the Membrane Filtration Unit (MFU) so it is ready for immediate use

When you are in the field:

- Find a flat area! Always place kit on a firm surface where it is easy to work
- Work in the shade where possible!
- Use the work surface in the kits - sterilise it before commencing and repeat as necessary to avoid cross-contamination
- After sterilisation of the MFU, rinse the sample cup 3 times with the sample to remove any traces of methanol
- Remember: The MFU apparatus needs to be sterilised **BEFORE** you take each new sample - not just prior to the first sample
- Remember: After preparing samples for incubation, label each petri dish with the relevant information - sample no./time/ source name etc. using the pen provided
- Remember: If collecting the sample in a bottle, store below 4ºC and analyse within 4-6 hours
- Remember: Switch the incubator on 30 minutes before use to allow time for incubation temperature to be reached. The incubator will show **'warming up'** during this initial period
- Remember: At least one hour and no more than four hours for resuscitation periods

Incubator Fault Codes:

Error 100: Temperature

The temperature of the incubator is outside the acceptable limit for reliable operation. Ensure the incubator is not located in extremely hot or cold conditions. Either **'Accept'** the fault to continue with the incubation cycle or select **'Back'** to cease the current cycle and restart when the conditions are appropriate.

Error 102: Low Temp

The incubator has failed to reach the temperature set-point in the time permitted. Ensure the conditions are suitable for incubation (no strongly cooling conditions are present) and either **'Accept'** to continue or **'Back'** to cease, relocate and restart.

Error 103: Over Temp

Temperature is too high for effective resuscitation or Peltier cooler is not reducing temperature sufficiently. Remove from direct sunlight if required and check air vent for blockages.

Error 107: Validation

A significant difference in measured response has occurred between the two temperature monitoring thermistors. The laser-trimmed thermistors are factory calibrated and designed for long-term field use. Any failure in the validation process indicates a hardware issue and the incubator should be returned to your local representative for attention. Select **'Back'** to accept the condition and continue operation until service can be arranged.

Error 110: Battery Low

The incubator will determine the power required at the start of an incubation and compare to currently available capacity. If the capacity is less than the power required the warning will be displayed. Select **'Accept'** to continue with the incubation cycle but find an additional power source to avoid losing power during microbiological analysis. Select **'Back'** to change power source prior to starting incubation.

Error 111: Battery Critical

Battery has 10 minutes life remaining. Find a replacement power source immediately. Select **'Back'** to stop incubation until more reliable supply is available. The message will remind at 1 minute intervals until a new power supply is attached.

Error 112: Power Loss

The message will be shown on start-up of the incubator if an unexpected loss of power occurred during an incubation cycle. Select **'Accept'** if the root cause has been determined.

Error 114: Overheat

Incubator is above acceptable temperature for safe operation. Select **'Back'** and check air vents for obstructions or remove from any source of indirect or direct heat. Select **'Accept'** if the condition is temporary and the incubator will be moved to a more acceptable location immediately.

Error 115: Critical Overheat

The message will be shown on start-up of the incubator if an automatic shut-down occurred due to a temperature being measured with potential to damage the incubator electronics. Check for causes such as blockages in air vents or direct/indirect heat and ensure condition does not remain.

Error 116: Thermistor Failure

The two independent temperature measurement thermistors will measure similar values in normal circumstances. If severe mechanical damage has occurred, the difference between the thermistor responses will be outside of specification and temperature display may be erratic/unexpected. Return the unit to your local representative for attention.

Incubator Use and Service

Should any condition arise requiring maintenance or service, please contact your local Palintest representative or via **support@palintest.com**

The **Potakit +** incubator is designed to provide field or static incubation of microbiological cultures at 37 or 44°C. Alternative uses will potentially expose the user to danger and negate warranty.

Potakit™ + Kit Contents

- Wagtech Incubator
- High performance Lead Acid battery
- Mains charger with international adapters, vehicle socket battery power lead, crocodile clip power leads
- Petri dish rack
- 20 Aluminium re-usable petri dishes
- Membrane Filtration assembly including bronze disc
- Pistol grip vacuum pump with silicone tubing
- 5 Media Measuring Devices (MMDs)
- 38.1g Membrane Lauryl Sulphate Broth
- 5 Pasteur pipettes
- Pen
- Hand lens
- Forceps
- 200 sterilised and sealed membrane filters
- 200 absorbent pads
- Absorbent pad dispenser
- Steel sampling cup with inert sampling cable
- Polypropylene 250ml beaker
- Sterilisable integrated work surface
- Cuvette brush
- 2 Dilution tubes
- Crush/stir rods
- De-ion pack
- Instructions
- Quick start prompt cards
- Double length turbidity tube
- Contour comparator, discs and cuvettes
- Comparator reagents for 200 tests for Free and Total Chlorine, Ammonia, Nitrite and Nitrate
- Pocket pH Sensor and pH calibration buffers
- Pocket Conductivity Sensor with conductivity calibration standard
- **VCDK** - Visual Arsenic Detection Kit, with consumables for 100 tests

Potakit + Incubator

Test Protocols	37 and 44°C temperature selections, user selectable time period, automatic resuscitation period option
Temperature Stability	±0.5°C
Temperature Control	Laser-trimmed thermistor pair with automatic temperature validation
User Interface	On screen and audible prompts available in English, French, Spanish and Chinese
Data Log	Last five incubation cycles report, view up to 100 incubation cycles via Wagtech Link app
Connectivity	Micro-USB connection to Windows devices
Size	80 x 60 x 260mm
Weight	400g
Power Supply	Sealed Lead Acid battery:12V DC/2A/30VA Mains power adapter: 100-240V AC/50-60 Hz/500mA/50VA. Vehicle and external battery connections provided
Power Consumption	High thermal efficiency heating system, 5 full incubation cycles from fully charged battery under standard conditions
IP Rating/Protection	Not specified. Do not immerse or clean using aggressive or corrosive cleaning agents. Do not steam clean the incubator
Humidity Rating	Up to 90% RH at 35°C (MIL-STD 810G)
Temperature Rating	0-50°C
Maximum Sound Pressure	EN61010-1, clause 12.5.1. A-weighted limit 79dBA, user prevented from setting higher volume

The **Potakit™ +** incubator is designed for incubation of microbiological samples in petri dishes. Follow the instructions regarding connection of power cables carefully and only use genuine Palintest cables/chargers. Using the **Potakit™ +** incubator in a manner not specified by Palintest may affect safe performance and invalidate warranty.

Part Number	Description
PTW 10452	Membrane Lauryl Sulphate Broth, 500g
PTW 10454	Membrane Lauryl Sulphate Broth, 38.1g
PTW 10456	1.92g Membrane Lauryl Sulphate Broth, pack of 25
PTW 10459	Membrane Filters, 47mm diameter, 200 pack
PTW 10460	Absorbent Pads and Membranes, 200 pack
PTW 10461	Membrane Filters, 47mm, 1000 pack
PTW 10462	Absorbent Pads and Membranes, 1000 pack
PTW 10463	Absorbent Pads, 100 pack
PTW 10464	Absorbent Pad Dispenser
PTW 10450	Coliform Starter Pack, includes absorbent pads, membranes and MLSB for 200 tests
PTW 10404-20	Aluminium Petri Dishes, 20 pack
PTW 10429	Media Measuring Devices, 5 pack
PTW 10430	Media Measuring Devices, 400 pack

Optional NutriDisks, Prepared Media and Spares

Part Number	Description
PTW 10060	NutriDisk pack for Faecal Streptococci, 100 pack
PTW 10062	NutriDisk pack for Pseudomonas aeruginosa, 100 pack
PTW 10064	NutriDisk pack for E. coli and Faecal Coliforms, 100 pack
PTW 10065	NutriDisk pack for Total Coliforms and E. coli, 100 pack
PTW 10066	NutriDisk pack for Total Colony Forming Units, 100 pack
PTW 10067	NutriDisk pack for Salmonella Typhi, 100 pack
PTW 10068	NutriDisk pack for E. coli, 100 pack
PTW 10069	NutriDisk pack for E. coli and coliforms, 100 pack
PTW 10468	Faecal Coliform Ampoules, 2.2ml
PTW 10470	Total Coliform Ampoules, 2.2ml
PTW 10410	Sterile Plastic Petri Dishes, 700 pack
PTW 10428	Wagpac Disposable Water Sample Bags
PTW 10446	Autoclave, Sterilisable, Portable, Economy Model
PTW 10425	Replacement 12V DC battery and cables, 8.5Ah, for Potakit + Incubator
PTW 10401	Pistol Grip Hand Vacuum Pump
PTW 10402	Bronze Disk
PTW 10403	Silicone Tubing for MFU, 6mm OD
PTW 10404	Sample Cup
PTW 10405	MFU/Silicone Tubing Connector
PTW 10412	Forceps
PTW 10416	Hand Lens
PTW 10700	Pasteur Pipettes, 1ml, pack of 5
PTW 19884	Pen
PT 500	De-ion Pack

Part Number	Description
AKW 152	Ammonia, 200 tests, Polypropylene Carton
AKW 179	Fluoride, 200 tests, Polypropylene Carton
AKW 031	Free and Total Chlorine (DPD 1 & 3), 250 tests, Polypropylene Carton
AKW 163	Nitrate (Nitratest), 200 tests, Polypropylene Carton
AKW 109	Nitrite (Nitricol), 200 tests, Polypropylene Carton
AK 152	Ammonia, 200 tests, Refill Pack
AK 179	Fluoride, 200 tests, Refill Pack
AK 031	Free and Total Chlorine (DPD 1 & 3), 250 tests, Refill Pack
AK 163	Nitrate (Nitratest), 200 tests, Refill Pack
AK 109	Nitrite (Nitricol), 200 tests, Refill Pack
PT 663	Cuvette Brush
PT 502	Crush/Stir Rods, Pack of 10
PT 512	Dilution Tube/Sample Tube
66104180	pH 4 buffer solution, 50ml
66104181	pH 7 buffer solution, 50ml
66104183	pH 10 buffer solution, 50ml
PT 142/7	Conductivity solution, 60ml
PTW 10438	2 Part Turbidity Tube
PT 521/5	Comparator Cuvettes, pack of 5
CKD 1001	Chlorine Comparator Disc, 0 - 5mg/litre Cl_2
CKD 1003	Chlorine Comparator Disc, 0 - 1mg/litre Cl_2
CKD 1109	Nitrite Comparator Disc, 0 - 0.4mg/litre N
CKD 1152	Ammonia Comparator Disc, 0 - 0.1mg/litre N
CKD 1163	Nitrate Comparator Disc, 0 - 15mg/litre N
CKD 1179	Fluoride Comparator Disc, 0 - 1.5mg/litre F

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