

## OPERATING INSTRUCTIONS

### ELE Paqualab System

418-160 Std System 50  
418-150 Std System 25  
418-100 Std System 25L

#### Applicable to:

**420-035** : Paqualab Incubator System 50  
**420-030** : Paqualab Incubator System 25  
**420-020** : Paqualab Incubator System 25L

#### Also including Operating Instructions for:

**430-550** : Paqualab Photometer

**430-020** : Paqualab pH/mV Meter

**513-160** : Paqualab Conductivity/TDS Meter

**430-260** : Paqualab Turbidity Meter

<p><b>ELE International</b> Chartmoor Road, Chartwell Business Park, Leighton Buzzard Bedfordshire, LU7 4WG England Sales: Civil +44 (0) 870 777 7706 Sales: Env. +44 (0) 870 777 7717 Service: +44 (0) 870 777 7727 fax: +44 (0) 1525 249249 email: <a href="mailto:ele@eleint.co.uk">ele@eleint.co.uk</a> <a href="http://www.ele.com">http://www.ele.com</a> <small>ELE International, a division of Danaher UK Industries Ltd</small></p>	<p><b>Distributor:</b></p>	<p><b>ELE International</b> Soiltest Product Division PO Box 389 Loveland, CO 80539 USA phone: +1 (800) 323 1242 fax: +1 (970) 663 9781 email: <a href="mailto:soiltest@eleusa.com">soiltest@eleusa.com</a> <a href="http://www.eleusa.com">http://www.eleusa.com</a> <small>ELE International, a division of Danaher Corporation</small></p>
<p><i>In the interests of improving and updating its equipment, ELE reserves the right to alter specifications to equipment at any time <b>ELE International 2005</b> ©</i></p>		

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**Part I : Daily Checklist of Equipment and Consumables**

**Item**

- 1 Carrying case with incubator
- 2 Aluminium petri dishes and carrier(s)
- 3 Mains connector and DC connector(s)
- 4 Aluminium vacuum flask, sample cup and cable
- 5 Aluminium filter funnel and collar
- 6 Aluminium filter assembly base
- 7 Upper and lower O rings (\*)
- 8 Bronze membrane support disc (\*)
- 9 Stainless steel forceps (\*)
- 10 Suction pump
- 11 0 – 60°C thermometer
- 12 Magnifying glass

**Consumables**

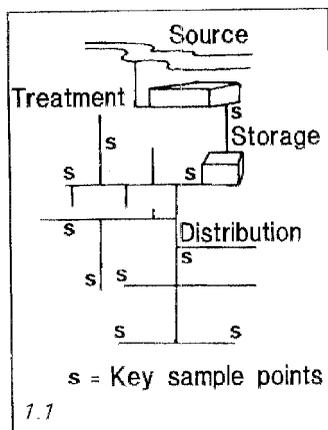
- 11 Membrane filters (\*)
- 12 Pads and dispenser (\*)
- 13 Membrane broth in polypropylene bottles (supplied in powdered form) (\*)
- 14 Operating instructions (\*)

(\*) These items are easily lost; always carry spares, which are available from ELE (follow the checklist every day)

**Part II : Planning and Support for Water Testing**

**1 Sampling programmes: selection of sites and frequency of sampling**

- 1.1 Samples should be taken from locations which are representative of the water source, treatment plant, storage facilities, distribution network and household connections. Where there are several sources and a mixed distribution system, it is necessary to construct the sampling programme to take account of this. Where there is a network distribution system, samples should be taken at random points evenly spread throughout the system. But where there are main branches and a remote periphery (as shown) greater attention should be devoted to the main branches and remote points in the distribution network.



- 1.2 In large urban populations, samples should be taken from the distribution system at the rate of at least one sample per 5000 of the population per month. For small urban and rural populations please refer to the following table.

Population served	Number of samples to be taken from treatment plant	Number of samples to be taken from distribution	Maximum sampling interval
Less than 1,000	1 per quarter	4 per quarter	3 months
1,001 to 2,000	1 per quarter	6 per quarter	3 months
2,001 to 3,000	1 per month	4 per month	1 month
3,001 to 5,000	1 per month	6 per month	1 month
5,001 to 10,000	1 per month	11 per month	1 month
10,001 to 20,000	2 per month	22 per month	2 weeks
20,001 to 30,000	2 per month	34 per month	2 weeks
30,001 to 50,000	4 per month	60 per month	1 week

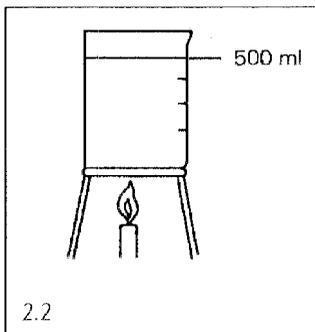
## 2 Preparation of bacteriological media from dehydrated powdered broth

### 2.1 *In a central laboratory*

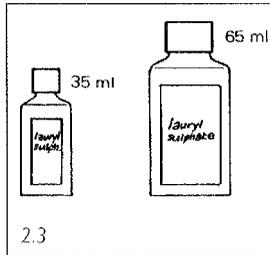
Weigh 38.1 grams membrane lauryl sulphate broth and add to 500 ml distilled water in a conical flask or beaker.

Alternatively add contents of one 4 test sachet into 10 ml of filtered water.

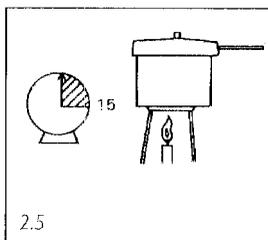
### 2.2 Heat the mixture with constant agitation until the powder is fully dissolved. Boiling should not be necessary.



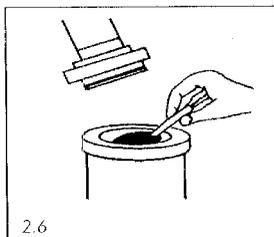
- 2.3 Dispense the medium into clean 35 ml or 65 ml polypropylene bottles. Ensure that there are no residues of previous medium or cleansing agent.



- 2.4 Replace bottle tops but leave them loose, **do not tighten**.
- 2.5 Autoclave bottles upright in a conventional autoclave (available from ELE International) at 121°C for 10 minutes, or place bottles in a pressure cooker and maintain steam at pressure for 15 minutes. Remove bottles, allow to cool, tighten tops and store in a cool, clean, dark environment.



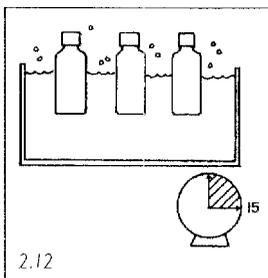
- 2.6 ***In the field***  
Place a membrane filter in the filtration assembly on the vacuum flask.



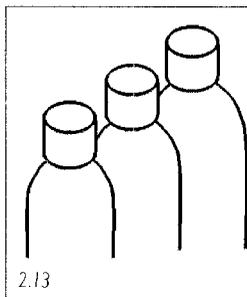
- 2.7 Select the cleanest water available, e.g. rainwater, sand-filtered water or, if necessary, stand raw water in a container overnight. **Do not use** water which has been chlorinated. Draw 500 ml water through the membrane (5 x 100 ml). If the water is turbid, more than one membrane may be required.
- 2.8 Remove the filtration assembly and check that the pH of the water is in the range 6.5 – 8.0. In the exceptional circumstance that the filtered water is not in this range, adjust the pH with sodium hydroxide solution or hydrochloric acid (dilute).
- 2.9 Add the full contents of a container of pre-weighed (38.1g) membrane lauryl sulphate broth to the 500 ml filtered water. If possible, heat to dissolve, e.g. place the vacuum flask and medium in a boiling water bath.

Alternatively add contents of one 4 test sachet into 10 ml of filtered water.

- 2.10 Dispense the dissolved medium into clean 35 ml or 65 ml polypropylene bottles.
- 2.11 Replace bottle tops and tighten firmly.
- 2.12 Place bottles containing medium in a boiler water bath for 15 minutes. Take care not to allow bottles to come into direct contact with the bottom of the water bath.

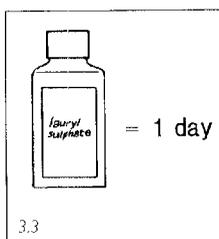


- 2.13 Remove bottles and store in a cool dark place before use. Use medium within 24 hours or repeat step 2.12 on two consecutive days. The medium may then be considered sterile and stored normally.



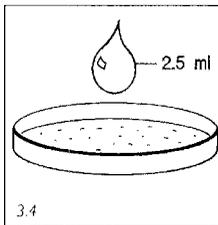
### 3 Notes on the field use of membrane lauryl sulphate broth

- 3.1 If stored at normal ambient temperature, the membrane broth should remain stable for several months. However, if there is any sign of deterioration, turbidity, yellowing or other denaturation, the contents of the bottle should be discarded.
- 3.2 Always clean empty media bottles thoroughly before re-use. Wash in hot water, if necessary with a little detergent; rinse several times in clean water, allow to dry and store dry in a clean environment with the tops replaced.
- 3.3 Ideally, only use a bottle of medium for one day's sampling. Do not try to make the contents of a bottle last over several days since this increases the chance of contamination.



- 3.4 Pour a slight excess of medium (2.5 ml approx) directly and carefully onto the sterile broth pads in the aluminium petri dishes before sampling and immediately re-close the medium bottle. Do not allow the top of the medium bottle to come into contact with the surfaces. This procedure may be performed in advance of the day's sampling programme, for example by dispensing the medium into 10 or 12 petri dishes before leaving base.

This has the advantage of reducing the number of manipulations in the field, but in some circumstances may be wasteful if the sampler is unsure of the number of samples to be processed that day. Pour off any excess medium before processing the sample, but make sure that the pad is **saturated**.

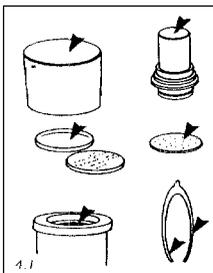


- 3.5 If only a small volume of broth medium is used from a bottle which is then immediately re-sealed, the medium may be re-sterilised by boiling in a water bath for 15 minutes.
- 3.6 Pre-prepared media is available in plastic ampoules which contain enough broth for one test. These ampoules have a shelf life of 1 year and should be stored in a cool dark place until needed.

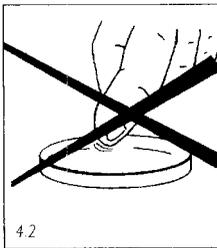
Undo top of ampoule and carefully squeeze base pouring medium into the base of a petri dish then add the absorbent pad. This ensures a more even distribution of medium than adding broth to a dish already containing the pad.

Impregnated pads contain membrane lauryl sulphate medium which is rehydrated using 2.5 ml of sterile water. If sterile water is not available, take the best quality non-chlorinated water and sterilise in a steam steriliser or boiling water bath. The pad is now ready for use.

#### 4 Notes on general hygiene in the field and sterilisation of equipment



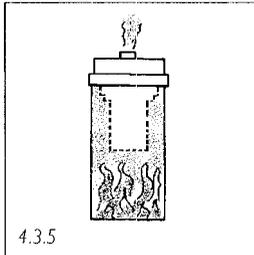
- 4.1 The most important surfaces which must remain clean and sterile are:
- 4.1.1 All those in contact with the water sample during processing, i.e. the inner surface of the aluminium sampling cup, the inner surface of the filter funnel and the upper surface of the filter base and membrane support.
  - 4.1.2 All those in contact with the broth medium, i.e. the inner surfaces of the aluminium petri dishes and the broth pad itself.
  - 4.1.3 All those in contact with the membrane, i.e. the lower filtration assembly, the broth pad and the forceps.
- 4.2 Outside surfaces may be handled but every care should be taken to restrict the time for which the internal surfaces are exposed to the environment. Under no circumstances should any of the above mentioned surfaces be exposed to direct contamination, e.g. by handling or splashing or from wind-blown material.



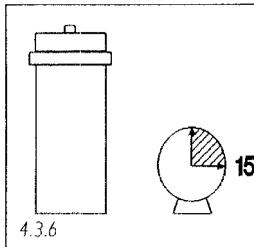
- 4.3 The aluminium sample cup and the filtration assembly is sterilised either immediately before or immediately after sampling, allowing the assembly to be transported sterile or ready for sterilisation.
- 4.3.1 Dry the sample cup and all components of the filtration assembly thoroughly using a clean cloth or paper tissue.
  - 4.3.2 Introduce 1.0 ml (approx) methanol into the sample cup and swirl around in the base of the cup.
  - 4.3.3 Carefully light the methanol using a taper, match or lighter. Avert the sample cup away from the face.
  - 4.3.4 Set down the sample cup, allow the methanol to continue burning for a few seconds and, while still alight, carefully invert the filtration assembly into the sample cup.

- 4.3.5 The methanol flame burns in the absence of sufficient oxygen forming formaldehyde vapour which permeates the entire assembly and completes the sterilisation process.

**Note:** the funnel and filter holding block should be assembled loosely so that the formaldehyde vapour can permeate the entire assembly.



- 4.3.6 At least fifteen minutes contact time must elapse before using the sterile sampling cup and filtration assembly. We recommend that sterilisation takes place after processing the sample, thereby leaving the sterile apparatus in place during transport to the next sampling point.



- 4.3.7 In some circumstances it is useful to have two filtration units so that one can be used whilst the other is being sterilised. There is space to accommodate two filtration units in the Paqualab 50 and Paqualab 25 systems.

**Note:** the use of excess methanol will result in a residue remaining in the sample cup after the sterilisation process has been completed. Experience will indicate the appropriate volume of methanol to introduce and the flaming time to avoid residues remaining in the sample cup.

- 4.4 The aluminium petri dishes should be cleaned carefully after use. Contaminated material should be destroyed by disinfection, burning or autoclaving. The dishes should be washed in hot water, if necessary with a little detergent, rinsed several times in clean water and dried. At this stage it may be appropriate to dry the inside surfaces of the dishes by exposure to direct sunlight. Provided that material does not enter the dishes during this process, the method gives a good degree of disinfection.
- 4.5 To sterilise the petri dishes, place 0.5 ml (approx) methanol in the base of the dish. Carefully light the methanol using a taper, match or lighter. Allow the methanol to almost burn out before placing the lid of the dish onto the base. Alternatively, boil both bases and lids in a water bath for 15 minutes. Drain off the water and whilst the dishes are still hot and dry, join bases with lids and stack. Alternatively, place complete dishes in a conventional oven at a temperature of 300°C for 30 minutes or autoclave at 121°C for 10 minutes, dry and stack.

## **5 Care of Equipment**

### **5.1 *Universal and Standard incubators***

A coiled mains lead for use with AC supplies is provided together with a twin terminal lead with crocodile clips for use with an external 12/24 V DC battery source e.g. car or motorcycle battery.

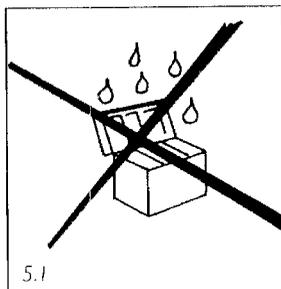
It is recommended to maintain the internal battery (Universal incubator) in a fully charged state. This helps preserve battery life.

**Always** incubate samples with the top of the kit closed and the unit placed away from draughts.

**Always** recharge the battery if the incubator section has been without a mains electricity source.

**Always** leave the battery fully charged if the water testing kit is to be stored or left unused for a few days, **i.e. leave the incubator connected to mains with the power switch off to maintain a trickle charge to the battery.**

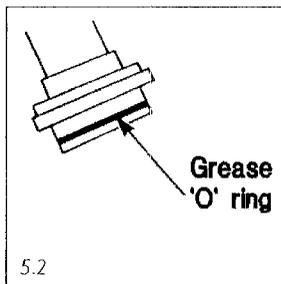
**Always** avoid the entry of water into the base of the water testing kit. The components are protected, but in humid environments it should not be assumed that they will resist corrosion indefinitely. Dry any spillages of water which occur **immediately.**



### 5.2 **Filtration apparatus**

Always dry the filtration apparatus, funnel, membrane support, filter base and vacuum flask thoroughly at the end of the day. This avoids the build-up of an oxide layer on the aluminium components.

Every 5 days, clean and dry the filtration apparatus. Smear a small quantity of silicone grease around the rubber 'O' rings which seal the components.

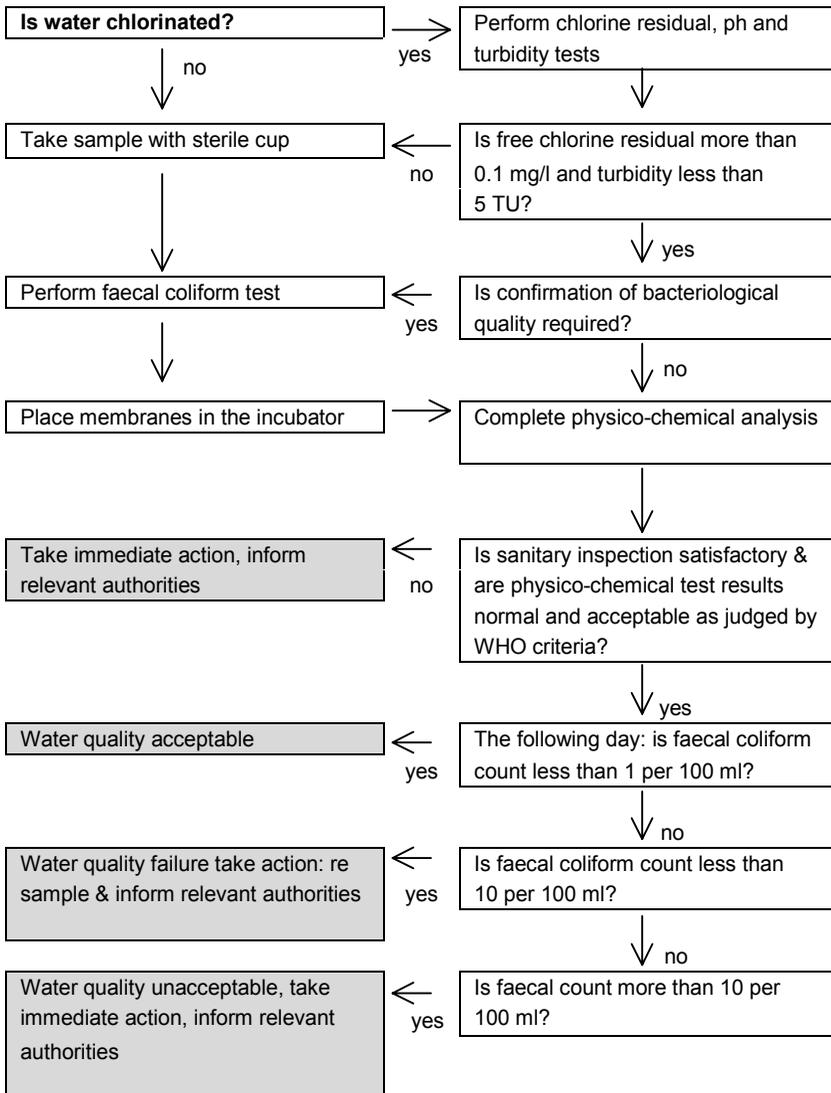


### 5.3 **Paqualab case**

The case is robust and resilient. It will tolerate a certain amount of rough handling, but every effort should be made to protect the box from abrasion, falls or other impacts.

**Part III : Processing**

**6 Checklist for the Operation of the Paqualab System**

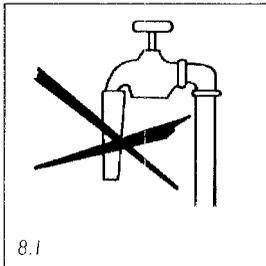


## **7 Sampling procedures: physico-chemical and bacteriological testing**

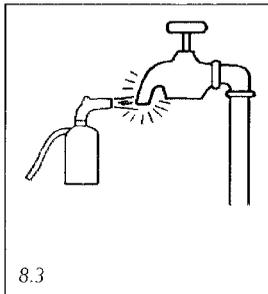
- 7.1 In the case of non-chlorinated supplies, the first test to be performed is the faecal coliform test. In these circumstances the first sample should be taken using the sterile sample cup. The bacteriological analysis is completed and then all physico-chemical tests are performed using the water remaining in the sample cup, if necessary supplementing the volume by re-sampling.

## **8 Sampling from a tap or similar outlet, e.g. tubewell handpump**

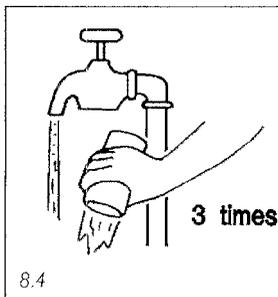
- 8.1 Remove all external fittings from the tap, e.g. anti-splash device, hose or nozzle. Check that there is no leakage from seals or fittings around the tap which might contaminate the sample. If present, leaks should be reported and repaired before bacteriological sampling.



- 8.2 Using a clean cloth, carefully clean the outlet of the tap, removing any loosely attached material or grease. Turn the tap on full and allow water to run to waste for one minute. This flushes the outlet of the tap and discharges water which has stood for a period in the service pipe.
- 8.3 Sterilise the tap using the flame of a blowlamp or gas torch until the whole tap is unbearably hot to the touch. This may take 20 to 30 seconds. Be careful not to flame seals and other non-metallic fittings directly. Alternatively ignite a piece of cotton wool or other absorbent material soaked in methylated spirit and hold it with a pair of tongs close to the outlet of the tap. Alternatively, place a strong bleach solution in a cup and hold it around the outlet of the tap for two minutes.



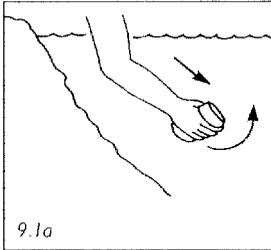
- 8.4 Allow the tap to flush by opening the tap to half speed and run to waste for 30 seconds. While the tap is running, place the clean vacuum flask in the gentle flow of water and rinse three times. **The flushing procedure prior to sampling should never be omitted.**



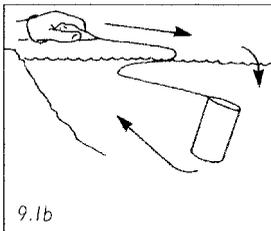
- 8.5 For non-chlorinated supplies run the tap to waste at half speed for 30 seconds, and place the sterile sample cup in the gentle flow of water until it is  $\frac{3}{4}$  full and carry out bacteriological analysis.

## 9 Sampling from a lake, reservoir or other surface water

- 9.1 Where there is good access to the surface water, it may be possible to sample by hand. In most circumstances it is inconvenient and undesirable to enter the water. Find a suitable place to lean over the water course and take the sample. Hold the sample cup firmly and plunge the open neck downwards into the water to a depth of 30 cm (approx). Turn the cup upwards and fill. Rinse the sample cup, re-sample and carry out bacteriological analysis. In locations with water flow, e.g. rivers and streams, the sample cup opening should face upwards.

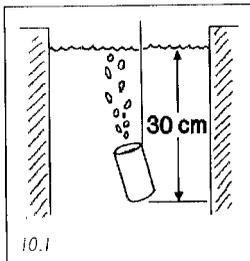


It is desirable to obtain the surface water sample as near as possible to the main body of water, e.g. in the main flow of a stream, not too close to the edge where water may be still unrepresentative. Furthermore, care must be taken not to introduce floating matter or material from the edge of the water course into the water sample. For this reason it may help to attach the sampling wire to the cup and take the sample from a bridge or other overhanging point. Alternatively the cup may be cast into the water from the edge and pulled slowly and carefully back towards the sampler.



## 10 Sampling from an open well or storage tank

- 10.1 Attach the sampling wire to the hole in the sample cup by means of the clip on the lower end of the wire. If necessary another length of wire or string may be attached to the first in order to increase the distance the sample cup may be lowered into the well or tank. Take great care not to lose the sample cup when sampling in this way.



- 10.2 Lower the sterile sample cup into the tank or well taking care not to allow it to come into contact with the walls of the structure. Allow the cup to submerge into the water to a depth of 30 cm (approx).
- 10.3 Raise the sample cup, again taking care not to allow it to come into contact with the sides of the well or tank.

## 11 Selecting the most appropriate volume for faecal coliform analysis

### 11.1 Contaminated water samples

The most appropriate volume of sample is that which will allow the most accurate enumeration of bacteria. This is achieved when the number of faecal coliform colonies on the membrane following incubation is in the range 20 – 200. If there are less than 20 colonies, there is the possibility of statistical errors. More than 200 colonies are usually difficult to count.

### 11.2 Treated drinking waters

For treated or distributed water samples it is hoped that the number of faecal coliform bacteria in the water is zero or minimal. Thus a preferred sample volume is set at 100 ml, and a count of zero faecal coliform bacteria per 100 ml is indicative of a microbiologically safe water supply.

### 11.3 All waters

Selection of sample volume is usually determined in the light of previous experience for a given source, treatment plant or distribution system. Drinking waters should be analysed using the Standard volume of 100 ml. If counts exceed 200, the source is grossly contaminated and action is urgently indicated.

There is little point in trying to compensate for such high results by lowering the sample volume. Take action. In fact, the same instruction applies to any drinking water where the result exceeds 1 faecal coliform per 100 ml (chlorinated supplies) or 10 faecal coliform per 100 ml (non-chlorinated supplies).

However, for source waters and partially treated waters (including those which are groundwater derived) it is valuable to adjust the sample volume to obtain faecal coliform counts in the optimum range 20-200. Typical volumes which may be appropriate for various water types are tabulated below. They are only guidelines; there is no substitute for experience of a given source. It may be useful to process more than one volume on the first occasion a water is sampled. In such cases it is not necessary to re-sterilise the filtration equipment between different volumes of the same sample, provided that the smaller volume is processed first.

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**Range of water sample volumes for faecal coliform analyses using the membrane filtration method for different waters**

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Sample type	Approximate volume		
	100 (ml)	50 (ml)	10 (ml)
1 Lakes, reservoirs, other surface water sources	*	**	***
2 Wells, boreholes, other protected sources	*	**	*
3 Water treatment plant – partially treated	**	**	*
4 Water treatment plant - treated	***		
5 Distribution system	***		

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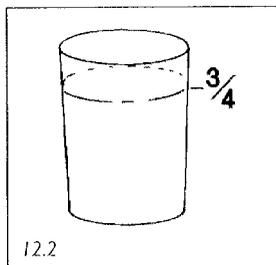
\*\*\* Normal volume of first choice  
\*\* Likely volume  
\* Possible volume

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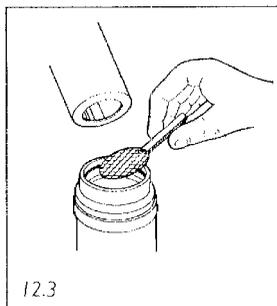
## 12 Processing samples for faecal coliform analysis in the field

- 12.1 Remove the sterile sample cup from the filtration assembly. Place filtration assembly upright on the vacuum flask and place the apparatus on a clean flat surface.

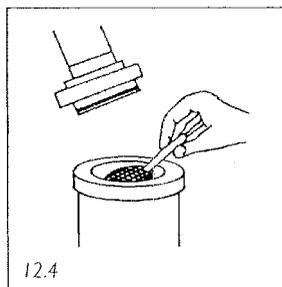
- 12.2 Taking great care not to introduce extraneous material into the sample cup, immediately take the sample, rinse the sample cup once and refill the sample cup to  $\frac{3}{4}$  full.



- 12.3 Loosen the filter funnel from the base support. Do not remove. Using flamed blunt-ended forceps, carefully remove a membrane filter from its sterile packaging having peeled back the transparent front cover.

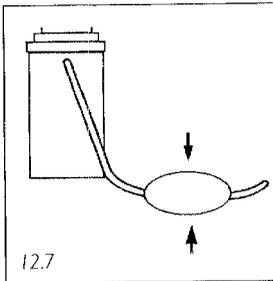


- 12.4 Holding the membrane only by the edge, place it grid side uppermost centrally on the sintered bronze membrane support. It should only be necessary to remove the aluminium filter funnel momentarily to do this.

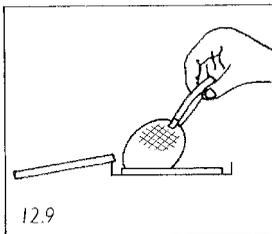


Take care not to tear the membrane. If it comes into contact with any surface other than the forceps or membrane support, or is damaged in any way, discard and start again.

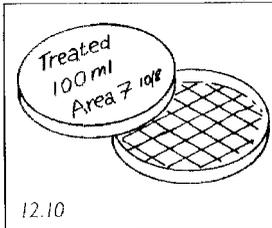
- 12.5 Place the forceps in a position where the tips cannot come into contact with contaminating surfaces. Lock the membrane in place by pushing the filter funnel down firmly.
- 12.6 Pour the sample up to the selected mark on the inside of the filter funnel (100 ml or 50 ml) ensuring that no other material enters the funnel.
- 12.7 Connect the vacuum pump to the filter base and draw a vacuum so that the sample passes quickly through the membrane. Release the vacuum by disconnecting the pump.



- 12.8 Taking the sterile forceps in one hand, loosen the filter funnel and remove. Lift the membrane filter out of the filter base by gripping the outer edge with the forceps tips.
- 12.9 Avoiding any contact with external surfaces, place the membrane, grid side uppermost, on a pad soaked in excess broth medium in a re-usable aluminium petri dish.



- 12.10 Ensure the absence of air bubbles between the membrane and broth pad. Replace the lid of the petri dish and label the top of the dish using waterproof marker with the sample code number and sample volume.



- 12.11 Place the petri dish, lid uppermost, at the bottom of the petri dish carrier, and place the carrier and petri dish stack in the incubator and replace lid. Subsequent sample dishes should be stacked down above the first in ascending order.
- 12.12 When the last sample of the day has been processed, wait a minimum of 60 minutes (resuscitation time) and then switch on the incubator. Try not to plan the day so that the period separating the first and last samples exceeds 5 hours. This restricts the maximum resuscitation period to 6 hours plus one hour (approx) warming time.

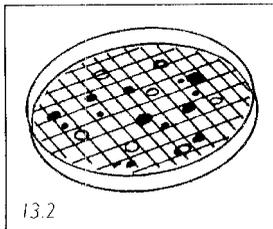
**Note:** resuscitation is particularly important for chlorinated or marine water samples where the faecal coliforms will be physiologically damaged or “stressed”. Try and maintain a delay of 3 hours before switching on the incubator for samples of this type. For faecal coliform analysis, incubate the samples at  $44^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$  for 12-16 hours. Switch off at the end of the incubation cycle. For total coliform analysis incubate the samples at  $37^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$  for 12-16 hours.

### 13 Counting faecal coliform and total coliform numbers, and recording results

- 13.1 Following incubation, remove the petri dish carrier from the incubator section. Remove the dishes from the carrier and place those containing membranes on one side. Taking one dish at a time, remove the lid and view the surface of the membrane in oblique light. If necessary, use a magnifying lens.

### 13.2 *Membrane Lauryl Sulphate Medium*

Count all yellow colonies which are 1mm in diameter. Do not count pale yellow colonies which fade to translucent on cooling, or pink colonies which have not fermented the lactose in the nutrient broth medium. If there are large numbers of colonies present, count methodically along each horizontal line of squares. In this way it should be possible to record counts in the range 0-300 colony forming units (cfu) per sample.



13.3 Immediately convert the count to numbers per 100 ml and record the result as "faecal coliforms per 100 ml" on the record sheet.

### 13.4 *Other Media*

If using dual test media, the number of Total coliforms and Faecal coliforms can be counted on one dish. Total coliforms are all red and all blue colonies. Faecal coliforms are blue colonies.

13.5 M-Endo broth is an alternative medium for coliforms. Total coliforms grow into 1mm diameter dark red colonies with a greenish gold sheen.

13.6 M-FC broth is used for Faecal coliforms (E.Coli) which form blue colonies.

13.7 Slanetz and Bartely medium is used for the enumeration of Faecal Streptococci whose colonies are red in colour.

**Part IV : Incubators**

**14 Incubator operation**

**14.1 Universal incubator (1484D0001)**

The Universal incubator can be operated from an external AC or DC power supply or internal rechargeable battery. It is equally suited to both laboratory and field requirements. Its rechargeable battery ensures continuity of power if operated from an interrupted AC mains supply.

**14.2 Standard incubator (1484D0500)**

The Standard incubator is exceptionally lightweight and portable. It requires an external 12/24 V DC power supply which, in the field, is normally readily available from either a vehicle or motor cycle battery. Despite its relative simplicity the Standard incubator is designed and manufactured to meet the same high performance as the Universal incubator.

**14.3 Operation from AC mains (Universal incubator)**

Before connecting the Universal incubator to AC mains, first check that the mains voltage selector switch is set correctly (120/240 V). Then connect the Universal incubator to a mains outlet using the coiled lead provided. The mains plug must be fused (3 A). If the incubator rechargeable cell is in a discharged condition then it is advisable to leave the incubator connected to mains (power switch off) for 24 hours before use since internal heat is generated in charging the battery and this should be avoided during incubation.

14.4 The Standard incubator cannot be operated directly from AC mains. However, it can be connected to the 12 V DC output of the Universal incubator using the short incubator linking lead (420-440).

**14.5 Operation from external DC source (either incubator)**

Check that the voltage selector is in the correct position (12 V or 24 V). Use the DC power lead (supplied) to connect the DC power source. The normal termination (as supplied) is crocodile clips to clamp to battery terminals. A cigar lighter plug is also provided.

The Standard incubator may be either connected directly to the DC power supply or to the DC output of the Universal incubator using the short incubator linking lead.

**Caution:** when the Universal incubator is switched off do not leave the Standard incubator connected to the output of the Universal incubator. There is a low current drain (30 mA) which will eventually discharge the battery.

#### 14.6 **Operation from internal rechargeable cell (Universal incubator)**

The internal rechargeable cell is designed so that it can be left on permanent charge and acts purely as a standby power supply for the incubator in the advent of a power failure.

**Note:** the battery can only be recharged from mains since the voltage of a normal vehicle battery is insufficient to provide the charging requirements.

If the Standard incubator is connected to the 12 V output of the Universal incubator then the rechargeable battery will operate both incubators until external power can be resumed.

The incubator should be left connected to mains with the power switch off. This will ensure that the battery remains fully charged, ready for immediate use.

Before taking the incubator out into the field, disconnect the unit from the mains, select 44°C and switch the power on for approximately 1 minute. Check that the power light is continuous green and the heater light is continuous yellow. For a more precise check of battery readiness, with the heater power switch on, place an accurate voltmeter across the 12/24 V DC output socket. Check that the voltage is not less than 12.5 V. Then switch the heater power off.

#### 14.7 **Protection from cold and draughts**

Where the incubator is part of a Paqualab 25 or Paqualab 50 system it is recommended that in temperate climates or draughty conditions, the incubator should be placed in its carrying case with the head closed. The external power lead can be passed through the small hinged door at the rear of the case.

#### 14.8 **Electrical protection**

Each incubator has a 3 A thermal circuit breaker to protect against overload. If it is tripped, check for incorrect external wiring or short circuit, then push to reset. If it trips again, then there may be a suspect internal fault, contact ELE International for advice.

**Note:** from serial no. 4094 the circuit breaker has been replaced by a 5 amp quick acting fuse. This has the same function as the circuit breaker but responds quicker to any fault giving greater protection to the control board.

A 2 amp anti-surge fuse is incorporated in the 3 pin chassis plug, this provides protection for the mains power unit. A spare fuse is contained in a compartment in the fuse holder.

A 10 amp slow acting fuse is fitted in line with the internal connection to the rechargeable cell to prevent excessive current in the unlikely event of a short circuit inside the incubator unit. If this fuse does fail then the power LED will fail to illuminate when operating from internal battery.

Separate the upper from the lower part of the case, examine for visual signs of short circuit and replace the fuse.

#### 14.9 **Commencement of incubation cycle**

- 14.9.1 Connect incubator to power supply.
- 14.9.2 Samples will have been placed on petri dishes. Ensure that they have been given sufficient time to resuscitate (see 12.12.).
- 14.9.3 Select temperature (37 or 44°C) and switch heater power on.

**Caution:** the incubator should not be operated outside the following ambient temperatures:

For 44°C incubation: 10 to 41°C

For 37°C incubation: 10 to 34°C

In hot climates it is advisable to carry out incubation during the cooler temperatures of the night or to place the incubator in a refrigerator or cool box.

#### 14.10 **Indicators and warnings**

- 14.10.1 Power light green, power sufficient.
- 14.10.2 Power light not illuminated – either internal 12 V battery or external supply low/faulty. Rectify fault or recharge battery immediately.
- 14.10.3 Heater light continuous yellow, temperature is rising.
- 14.10.4 Heater light yellow, cycling on/off heater controlling at pre-selected temperature.

#### 14.11 **Incubator temperature control**

The incubator's temperature control system incorporates extremely stable ( $\pm 1^\circ\text{C}$ ) temperature reference and monitoring components.

Approximately 45 minutes after switching heater power on the heater LED changes from continuous yellow (incubator temperature still rising) to cycling on/off, indicating that the incubator block has reached the set temperature. A further hour should be allowed for the petri dish assembly to reach the same temperature as the surrounding block.

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Incubator temperature can be monitored approximately ( $\pm 1^{\circ}\text{C}$ ) by the use of an accessory kit comprising incubator bung with clearance hole together with a 0-60 $^{\circ}\text{C}$  spirit in glass thermometer (included with the Paqualab 25 and Paqualab 50 systems). However, for precise temperature measurement (e.g. when altering incubator temperature), a mercury in glass, partial immersion thermometer graduated 0.1 $^{\circ}\text{C}$  should be used (e.g. ASTM 91C which is recognised as an International Standard).

**Note:** the incorporation of a mercury in glass thermometer into the incubator has been deliberately avoided because of the restrictions applied to transportation of mercury by air.

**The procedure for accurately monitoring and adjusting incubator temperature is detailed in Appendix B.**

**Appendix A: Specifications**

**1484D0500 Standard Incubator, 1484D0001 Universal Incubator and  
420-450 Paqualab Filtration Unit**

**Features common to both Universal and Standard Incubators**

Construction	Aluminium housing. Wipe clean, white, epoxy polyester finish. Splits into two parts for ease of maintenance. Incubation chamber constructed from a hollow, cylindrical block of aluminium surrounded by a 25mm thick thermal insulation layer.
Sample dishes	Supplied with stack of 25 re-usable aluminium petri dishes, 55mm diameter, 4mm height. Will also accommodate up to 10 plastic petri dishes (Sterilin, Gelman or similar).
Heater power requirements (20°C ambient)	During the first hour after switch on approximately 13 watts. Thereafter, approximately 3 watts.
Incubator temperature	Switchable to either of two set temperatures in the range 20-50°C. Normally set to 37.0°C and 44.0°C.
Ambient temperature	Ambient operating range: 5°C up to 41°C or 3°C below the incubator temperature (whichever is the lower).
Incubator temperature	Ambient constant: within $\pm 0.2^\circ\text{C}$ of set temperature. Ambient varying accuracy between 10-35°C: within $\pm 0.5^\circ\text{C}$ of set temperature.
Temperature adjustment	User accessible adjustment screws behind access plate.
Temperature monitoring	<p>The incubator's temperatures control system incorporates extremely stable (<math>\pm 0.1^\circ\text{C}</math>) temperature reference and monitoring components. When the heater LED changes from continuous yellow (incubator still warming up) to cycling on/off, this indicates that the incubator block has reached the set temperature. A further hour should be allowed for the petri dish assembly to reach the same temperature as the surrounding block.</p> <p>Incubator temperature can be monitored approximately (<math>\pm 1^\circ\text{C}</math>) by the use of an accessory kit comprising incubator bung with clearance hole together with a 0-60°C spirit in glass thermometer, (included with Paqualab 25 and Paqualab 50 systems). However, for precise temperature measurement (e.g. when altering incubator temperature), a mercury in glass, partial immersion thermometer graduated to <math>0.1^\circ\text{C}</math> should be used (e.g. ASTM 91C which is recognised as an International Standard).</p> <p><b>Note:</b> the incorporation of a mercury in glass thermometer into the incubator has been deliberately avoided because of the restrictions applied to transportation of mercury by air.</p>
Safety features	Low voltage: power light not illuminated. DC supply polarity reversed: supply automatically disconnected. Short circuit or current overload: thermal cutout with reset.

**420-010 Standard Incubator** (incorporated in Paqualab 25L and 50 systems)

Dimensions	130 x 160 x 190 mm (length x width x height)
Weight	2 kg
Electric power requirements	<p>Either of the following external DC supplies:</p> <p>12 V DC, 1 A maximum          24 V DC, 0.5 A maximum</p> <p><b>Note:</b> either positive or negative ground is acceptable.</p> <p>The Standard incubator can also be powered from the 12/24 V DC outlet of the Universal incubator.</p>
Power leads and connectors supplied	External DC lead with crocodile connectors and vehicle cigar lighter plug. A short umbilical lead is also provided in order to power the Standard incubator from the Universal incubator 12/24 V DC output.

**420-015 Universal Incubator** (incorporated in Paqualab 25 and 50 systems)

Dimensions	220 x 160 x 190 mm (length x width x height)
Weight	6 kg (including 2.4 kg for rechargeable battery)
Electric power requirements	<p>Any of the following :</p> <p>12 V DC, 1 A maximum          24 V DC, 0.5 A maximum</p> <p><b>Note:</b> either positive or negative ground is acceptable.</p> <p>100/120 V, 50/60 Hz, AC mains          200/240 V, 50/60 Hz, AC mains</p> <p>Internal, rechargeable 12 V, 7 AH battery providing power back-up, if used with interrupted mains supply. If the ambient temperature is below 20°C then this capacity will be reduced). No air/sea shipping restrictions.</p>
Mains power circuit	Incorporates a high efficiency, cool running, switch mode design which permits operation at high ambient close to incubator temperature. The battery is automatically recharged when the incubator is connected to AC mains.
Power outlet	12/24 V outlet to supply Standard incubator, normally 12 V unless using external 24 V supply.
Power leads and connectors supplied	Spring-coiled mains lead. External DC lead with crocodile connectors and vehicle cigar lighter plug.

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**420-450 Paqualab Filtration Unit**

Dimensions	Individual components pack into a compact cylindrical package 120 x 80 mm (height x diameter)
Weight	0.66 kg
Construction	Main components made from anodised aluminium.
Sterilisation	Sterilisable in the field using anaerobic combustion of methanol.
Comprises	Sampling cup, complete with lanyard, support block incorporating 38mm sintered bronze disc to retain filter grid, filtration funnel with 50 ml and 100 ml graduated markings, sample receiving cup fitted with vacuum connection and hand-operated suction pump.

***Appendix B: Measurement and Control of Incubator Temperature***

The temperature monitoring and reference components used in the incubator temperature control circuit have long-term stability of  $\pm 0.1^{\circ}\text{C}$ . Consequently, once the incubator temperature has been set, it should only be necessary to check the incubator temperature if either a fault is experienced or if the incubator temperature is to be changed.

The incubator may be set to either of two pre-selected temperatures within the range 20.0-50.0 $^{\circ}\text{C}$ . The temperatures will normally be factory preset accurately to 37.0 $^{\circ}\text{C}$  and 44.0 $^{\circ}\text{C}$  respectively but can, alternatively, be preset to any two temperatures within the range 20.0-50.0 $^{\circ}\text{C}$ .

Each Paqualab 25 and Paqualab 50 system is supplied with an incubator bung with thermometer clearance hole together with a simple 0-60 $^{\circ}\text{C}$  spirit thermometer (accurate to  $\pm 1^{\circ}\text{C}$ ). This thermometer should only be used as an approximate guide (e.g. if a fault is suspected). Remove the lid of the uppermost petri dish and insert the thermometer through the clearance hole in the bung until the thermometer bulb comes into contact with the wet absorbent pad on the uppermost petri dish. Secure thermometer in position by pressing plasticine or modelling clay around the top of the bung. This check is best performed using a stack of approximately 12 petri dishes with the pads moistened with water (to simulate the humid incubation environment). However, in order to carry out accurate temperature measurement, a precision thermometer should be used, e.g. ASTM 91C (20-60 $^{\circ}\text{C}$  with 0.1 $^{\circ}\text{C}$  graduations, accurate to  $\pm 0.2^{\circ}\text{C}$ ). This is an internationally recognised Standard thermometer. ELE can supply such a thermometer, but it would be very expensive to freight owing to transport regulations governing mercury as a hazardous chemical. We therefore recommend local purchase.

Select the most appropriate incubator temperature, switch the heater power on and allow approximately 2 hours for the incubator temperature to stabilise. Locate the inspection plate on the side of the incubator and remove the single retaining screw. The temperature adjustment screws can then be seen. The left hand adjustment screw corresponds to the 37 $^{\circ}\text{C}$  switch position, and the right hand adjustment screw corresponds to the 44 $^{\circ}\text{C}$  switch position.

Adjust the appropriate screw either anti-clockwise to increase temperature or clockwise to decrease. One turn will cause approximately 1 $^{\circ}\text{C}$  change in the incubator temperature. Allow approximately  $\frac{1}{2}$  hour for the temperature to stabilise before re-checking.

Finally, monitor the temperature for 3 or 4 hours to ensure that it is stable.

**Appendix C: Paqualab Photometer Calibration Charts and Test Procedures**

**433-102 and 433-102/10 Ammonia**

*Test for Ammonia in natural drinking and waste water*

Ammonia occurs as a breakdown product of nitrogenous material in natural waters. It is also found in domestic effluents and certain industrial waste waters. Ammonia is harmful to fish and other forms of aquatic life, and the ammonia level must be carefully controlled in water used for fish farms and aquariums. Ammonia tests are routinely applied for pollution control on effluents and waste waters, and for the monitoring of drinking water supplies.

The Ammonia Test provides a simple method of measuring ammonia (ammonical nitrogen) over the range 0-1.0 mg/l N.

**Method**

The Ammonia test is based on an indophenol method. Ammonia reacts with alkaline salicylate in the presence of chlorine to form a green-blue indophenol complex. Catalysts are incorporated to ensure complete and rapid colour development. The reagents are provided in the form of two tablets for maximum convenience. The test is simply carried out by adding one of each tablet to a sample of the water.

The intensity of the colour produced in the test is proportional to the ammonia concentration and is measured using the Photometer.

**Reagents and Equipment**

Ammonia No. 1 Tablets  
Ammonia No. 2 Tablets  
Photometer  
Round Test Tubes, 10 ml glass

**Test Procedure**

- 1 Fill test tube with sample to the 10 ml mark.
- 2 Add one Ammonia No. 1 Tablet and one Ammonia No. 2 Tablet, crush and mix to dissolve.
- 3 Stand for ten minutes to allow colour development.
- 4 Select wavelength **640 nm** on Photometer.
- 5 Take Photometer reading in usual manner (see Photometer instructions).
- 6 Consult Ammonia Calibration Chart.

**Ammonia  
Calibration  
Chart**

%T	Ammonia			Ammonia mg/l N						640 nm	
	9	8	7	6	5	4	3	2	1	0	
80	-	-	-	-	-	0.00	0.00	0.01	0.01	0.02	
70	0.02	0.03	0.03	0.04	0.04	0.05	0.05	0.06	0.06	0.07	
60	0.07	0.08	0.09	0.09	0.10	0.11	0.11	0.12	0.13	0.13	
50	0.14	0.15	0.16	0.16	0.17	0.18	0.19	0.20	0.20	0.21	
40	0.22	0.23	0.24	0.25	0.26	0.27	0.28	0.29	0.30	0.31	
30	0.32	0.33	0.34	0.36	0.37	0.38	0.39	0.41	0.42	0.44	
20	0.45	0.47	0.48	0.50	0.51	0.53	0.55	0.57	0.59	0.61	
10	0.63	0.66	0.68	0.71	0.74	0.77	0.80	0.83	0.87	0.91	
0	0.96	1.00	-	-	-	-	-	-	-	-	

**Sea Water  
Samples**

Ammonia Conditioning Reagent is required when testing sea water or brackish water samples to prevent precipitation of salts. The reagent is supplied in a special "spoon pack" to aid measuring out the powder.

Fill the test tube with sample to the 10 ml mark, and add one level spoonful of conditioning reagent. Mix to dissolve reagent then continue the test as described in the above test procedures.

**Notes**

- 1 At low temperature the rate of colour development in the test may be slower. If the sample temperature is below 20°C allow 15 minutes for the colour to develop.
- 2 Ammonia concentrations can be expressed in a number of different ways. The following factors may be used for the conversion of readings:  
To convert from N to NH<sub>4</sub> multiply by 1.3.  
To convert from N to NH<sub>3</sub> multiply by 1.2.

### **433-115 and 433-115/10 Chlorine (DPD)**

*Test for Free, Combined and Total Chlorine in water*

Chlorine and chlorine-release compounds are widely used for the disinfection of drinking water and swimming pools, for the control of micro-biological growth in cooling water, and in many other water treatment systems. Accurate measurement of the chlorine residual is an essential aspect of the control of these chlorination processes.

The chlorine level can be expressed in terms of the free chlorine, combined chlorine or total chlorine residuals. For the majority of applications measurement of the free chlorine residual is the most important. The DPD chlorine method provides a simple means of measuring free, combined and total chlorine residuals over the range 0-5 mg/l.

#### **Method**

This Chlorine test uses the DPD method developed by Dr AT Palin and now internationally recognised as the standard method of testing for chlorine and other disinfectant residuals. In the DPD method the reagents are provided in tablet form for maximum convenience and simplicity of use.

Free chlorine reacts with diethyl-p-phenylene diamine (DPD) in buffered solution to produce pink colour. The intensity of the colour is proportional to the free chlorine concentration. Subsequent addition of excess potassium iodide induces a further reaction with any combined chlorine present. The colour intensity is now proportional to the total chlorine concentration; the increase in intensity represents the combined chlorine concentration. In this way it is possible to differentiate between free and combined chlorine present in the sample. The colour intensities are measured using the Photometer.

#### **Reagents and Equipment**

DPD No. 1 Tablets  
DPD No. 3 Tablets  
Photometer  
Round Test Tubes, 10 ml glass

#### **Test Procedure**

1 Rinse test tube with sample leaving two or three drops of sample in the tube.

- 2 Add one DPD No. 1 Tablet, crush tablet and then fill the test tube with sample to the 10 ml mark. Mix to dissolve tablet.
- 3 Select wavelength **520 nm** on Photometer.
- 4 Take Photometer reading immediately in usual manner – see Photometer instructions. Consult Chlorine (DPD) Calibration Chart.

**Chlorine  
Calibration  
Chart**

%T	Chlorine (DPD)			Chlorine mg/l				520 nm		
	9	8	7	6	5	4	3	2	1	0
90	0.01	0.02	0.04	0.05	0.06	0.07	0.08	0.10	0.11	0.12
80	0.14	0.15	0.16	0.18	0.19	0.21	0.21	0.22	0.24	0.25
70	0.26	0.27	0.29	0.30	0.32	0.33	0.35	0.36	0.38	0.40
60	0.42	0.44	0.46	0.48	0.50	0.51	0.53	0.55	0.57	0.59
50	0.61	0.63	0.65	0.68	0.70	0.72	0.74	0.76	0.78	0.80
40	0.82	0.85	0.87	0.89	0.91	0.93	0.96	0.98	1.00	1.03
30	1.06	1.09	1.13	1.17	1.20	1.24	1.28	1.31	1.35	1.38
20	1.42	1.46	1.51	1.56	1.64	1.70	1.77	1.84	1.92	2.00
10	2.08	2.16	2.24	2.32	2.40	2.50	2.60	2.72	2.84	3.00
0	3.20	3.40	3.70	4.00	4.50	5.00	-	-	-	-

- 5 The result represents the **free chlorine** residual as milligrams per litre. Stop the test at this stage if only free chlorine determination is required.
- 6 If it is desired to measure combined or total chlorine residual continue the test on the same test portion.
- 7 Add one DPD No. 3 Tablet, crush and mix to dissolve.
- 8 Stand for two minutes to allow full colour development.
- 9 Take Photometer reading. Consult Chlorine (DPD) Calibration Chart.
- 10 The result represents the total chlorine residual as milligrams per litre.

11 The **combined chlorine** residual is obtained by subtracting the free chlorine residual result from the total chlorine residual result.

i.e. Combined Chlorine = Total Chlorine – Free Chlorine

**Note**

A too high chlorine level (above 10 mg/l) can cause bleaching of the pink colour formed in the DPD test and give a false negative result. If a colourless test solution is obtained when chlorine is known to be present, check for the possibility of bleaching by repeating the test on a sample diluted with chlorine-free water.

### **433-166 and 433-166/10 Nitrate (Nitratest)**

*Test for Nitrate in Natural Drinking and Waste Water*

Nitrates are normally present in natural, drinking and waste waters. Nitrates enter water supplies from the breakdown of natural vegetation, the use of chemical fertilisers in modern agriculture and from the oxidation of nitrogen compounds in sewage effluents and industrial wastes.

Nitrate is an important control test for water supplies. Drinking waters containing excessive amounts of nitrates can cause methaemoglobinaemia in bottle-fed infants (blue babies). The EEC has set a recommended maximum of 5.7 mg/l N (25 mg/l NO<sub>3</sub>) and an absolute maximum of 11.3 mg/l N (50 mg/l NO<sub>3</sub>) for nitrate in drinking water.

The Nitratest method provides a simple test for nitrate nitrogen over the range 0-1 mg/l N. The test can however be extended to cover the range 0-20 mg/l by a simple dilution technique.

#### **Method**

In the Nitratest method nitrate is first reduced to nitrite, the resulting nitrite is then determined by a diazonium reaction to form a reddish dye.

The reduction stage is carried out using the unique zinc-based Nitratest Powder, and Nitratest Tablet which aids rapid flocculation after the one minute contact period. The test is conducted in a special Nitratest Tube – a graduated sample.

The nitrite resulting from the reduction stage is determined by reaction with suphhanilic acid in the presence of N-(1-naphthyl)-ethylene diamine to form a reddish dye. The reagents are provided in a single Nitricol tablet which is simply added to the test solution.

The intensity of the colour produced in the test is proportional to the nitrate concentration and is measured using the Photometer.

#### **Reagents and Equipment**

Nitratest Powder (Spoon Pack)  
Nitratest Tablets  
Nitricol Tablets  
Nitratest Tube, 20 ml  
Photometer  
Round Test Tubes, 10 ml

- Test Procedure**
- 1 Fill the Nitratest Tube with sample to the 20 ml mark.
  - 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 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 two minutes or longer 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 round test tube, filling to the 10 ml mark.
  - 5 Add one Nitricol tablet, crush and mix to dissolve.
  - 6 Stand for 10 minutes to allow full colour development
  - 7 Select wavelength **570 nm** on Photometer.
  - 8 Take Photometer reading in usual manner (see Photometer instructions).
  - 9 Consult Nitratest Calibration Chart.

**Nitratest Calibration Chart**

%T	Nitratest									
	9	8	7	6	5	4	3	2	1	0
90	-	-	-	-	.000	.003	.006	.009	.012	.015
80	.018	.021	.024	.028	.032	.036	.040	.043	.047	.051
70	.055	.059	.063	.068	.072	.076	.080	.085	.089	.094
60	0.10	0.10	0.11	0.11	0.12	0.12	0.13	0.13	0.14	0.14
50	0.15	0.15	0.16	0.17	0.17	0.18	0.18	0.19	0.20	0.20
40	0.21	0.22	0.22	0.23	0.24	0.24	0.25	0.26	0.27	0.28
30	0.30	0.31	0.32	0.33	0.34	0.35	0.37	0.38	0.40	0.42
20	0.43	0.45	0.47	0.50	0.53	0.55	0.58	0.60	0.65	0.70
10	0.75	0.80	0.85	0.90	0.95	1.00	-	-	-	-

To convert mg/l N to mg/l NO<sub>3</sub> multiply results by 4.4

Concentrations of nitrate greater than 1.0 mg/l may be determined by diluting the original sample with deionised water. The test can be conveniently carried out over a range 0-20 mg/l N as follows:

Take a clean Nitratest Tube. Add 1 ml of sample using a pipette or graduated dropper. Fill the Nitratest Tube to the 20 ml mark with deionised water. Continue the test procedure as given in steps 2 to 9 above. Multiply the chart reading obtained by 20 to obtain the nitrate concentration in the original sample.

***Nitrite  
Correction***

The Nitratest method will also respond to any nitrite present in the sample. In most natural and drinking waters the amount of nitrite will be small in comparison to the nitrate concentration. If it is desired to correct for nitrite, determine nitrite concentration (as mg/l N) in the prescribed manner and deduct from the nitrate concentration (as mg/l N) obtained from the Nitratest procedure.

**433-168 and 433-168/10 Nitrite (Nitricol)**

*Test for Nitrite in Natural Drinking and Waste Water*

Nitrites are found in natural waters as an intermediate product in the nitrogen cycle. Nitrite is harmful to fish and other forms of aquatic life and the nitrite level must be carefully controlled in water used for fish farms and aquariums. The nitrite test is also applied for pollution control in waste waters, and for the monitoring of drinking water.

The Nitricol test provides a simple method of measuring Nitrite Nitrogen levels over the range 0 to 0.5 mg/l N. Higher levels can be determined by diluting the sample.

**Method**

Nitrites in acid solution react with sulphanilic acid. The resulting diazo compound couples with N-(1-naphthyl)-ethylene diamine to form a reddish dye. The Nitricol method features a single tablet reagent containing both of these reagents in an acidic formulation. The test is simply carried out by adding a tablet to a sample of the water under test.

The intensity of the colour produced in the test is proportional to the nitrite concentration and is measured using the Photometer.

**Reagents and Equipment**

Nitricol Tablets  
Photometer  
Round Test Tubes, 10 ml glass

**Test Procedure**

- 1 Fill round test tube with sample to the 10 ml mark.
- 2 Add one Nitricol Tablet, crush and mix to dissolve.
- 3 Stand for 10 minutes to allow full colour development.
- 4 Select wavelength **520 nm** on Photometer.
- 5 Take Photometer reading in usual manner (see Photometer instructions).
- 6 Consult Nitricol Calibration Chart.

**Nitricol  
 Calibration  
 Chart**

	<b>Nitricol</b>		<b>Nitricol mg/l N</b>								<b>520 nm</b>	
	%T	9	8	7	6	5	4	3	2	1	0	
90	.000	.001	.003	.004	.006	.007	.009	.011	.012	.014		
80	.016	.018	.019	.021	.023	.025	.027	.028	.030	.032		
70	.034	.036	.038	.040	.042	.044	.046	.048	.051	.053		
60	.055	.057	.060	.062	.064	.067	.069	.072	.074	.077		
50	.079	.082	.084	.087	.090	.093	.096	.099	.102	.105		
40	.108	.111	.114	.118	.121	.124	.128	.132	.135	.139		
30	.143	.147	.151	.155	.160	.164	.169	.173	.178	.183		
20	.189	.194	.200	.205	.212	.218	.224	.231	.238	.246		
10	.254	.262	.271	.280	.290	.301	.312	.325	.338	.353		
0	.369	.387	.408	.431	.460	.500	-	-	-	-		

*To convert from mg/l N to mg/l NO<sub>2</sub> multiply result by 3.3.*

**433-180 and 433-180/10 pH (Phenol Red)**

*Test for pH Value of Water and Aqueous Solutions*

pH measurement is one of the tests most frequently carried out on water and aqueous solutions. The phenol red indicator method provides a simple colorimetric means of pH determination for neutral and slightly alkaline waters over the range 6.8-8.4 units.

**Method**

The Phenol Red test uses a tablet reagent containing the precise amount of phenol red indicator required for the test. Phenol red reacts in water at different pH values over the range 6.8-8.4 to produce a distinctive range of colours from yellow to red. The colour of the test solution is indicative of the pH value and is measured using the Photometer.

Phenol red tablets contain a dechlorinating agent so that the test can be carried out in water containing normal levels of chlorine or other disinfectant residuals.

**Reagents and Procedure**

Phenol Red Clear Tablets  
Photometer  
Round Test Tubes, 10 ml glass

**Test Procedure**

- 1 Fill test tube with sample to the 10 ml mark.
- 2 Add one Phenol Red Tablet, crush and mix to dissolve.
- 3 Select wavelength **520 nm** on Photometer.
- 4 Take Photometer reading in usual manner (see Photometer instructions).
- 5 Consult Phenol Red Calibration Chart (Transmittance-display photometer only).

<b>Phenol Red Calibration Chart</b>	<b>Phenol Red</b>		<b>pH Units</b>								<b>520 nm</b>		
	%T	9	8	7	6	5	4	3	2	1	0		
60	6.80	6.80	6.85	6.85	6.90	6.90	6.95	6.95	6.95	6.95	7.00		
50	7.00	7.05	7.05	7.10	7.10	7.15	7.15	7.20	7.20	7.20	7.20		
40	7.25	7.25	7.30	7.30	7.35	7.35	7.40	7.40	7.45	7.45	7.45		
30	7.50	7.50	7.50	7.55	7.55	7.60	7.60	7.65	7.65	7.70	7.70		
20	7.70	7.70	7.75	7.75	7.80	7.80	7.85	7.85	7.90	7.90	7.90		
10	7.95	8.00	8.05	8.10	8.15	8.20	8.25	8.30	8.40	8.40	-		

- Notes**
- 1 The colour range of the phenol red test is yellow, through orange, to red. The formation of an intense purple coloration shows that the indicator has been affected by high chlorine or other disinfectant residuals. In such cases the result should be disregarded.
  - 2 Phenol red does not show any further colour change at pH values below 6.8 or above 8.4. Note therefore that when such values are recorded this could indicate that the sample has a much lower or much higher pH value.
  - 3 Ionic strength, temperature and other water factors may have an effect on pH readings.

***Appendix D: Water Quality Daily Report Sheet***

**14 Report Sheet**

15.1 A blank Report Sheet is included. This can be detached from this manual and copied as required for note-taking in the field.

**Water quality  
daily report sheet**

Water Authority

Date

District

Sampler/Analyst

Sample number

1 Location

2 Source

3 Time

4 Physical  
description

5 Odour/taste

6 Turbidity (NTU)

7 Free chlorine  
residual (mg/l)

8 Combined  
chlorine (mg/l)

9 Conductivity  
( $\mu\text{S}/\text{cm}$ )

10 Temperature ( $^{\circ}\text{C}$ )

11 pH

12 Faecal coliforms  
(E.coli/100 ml)

13 Total coliforms  
(E.coli/100 ml)

14 Unsatisfactory results (+)

15 Recommended Action