

omniPAGE Maxi and Mini Wide Vertical Electrophoresis and Electroblotting Units

Instruction Manual

Catalogue Numbers

VS10WCBS

SB10W

VS20CBS

SB20

Record the following for your records:

Model _____

Catalogue No. _____

Date of Delivery _____

Warranty Period _____

Serial No. _____

Invoice No. _____

Purchase Order No. _____

Contents

Instruction Manual	1
Catalogue Numbers	1
Contents	2
Safety Information	3
Packing List	4
Specifications	5
Operating Instructions	6
Usage Guidance and restrictions	6
Setting up the omniPAGE Gel Tanks	6
Casting Unit Preparation	6
Gel Preparation	110
Sample preparation and Loading	13
Gel Running	15
Solutions (For SDS-PAGE)	16
References	17
Protein Blotting using the omniPAGE Electroblotter	27
Buffer Solutions (For Blotting)	270
Troubleshooting	20
Care and Maintenance	277
Cleaning Vertical Units	277
RNase Decontamination	277
Ordering information	28
Comb options	29
Related Products	30
Warranty	32

Safety Information



When used correctly, these units pose no health risk. However, these units can deliver dangerous levels of electricity and are to be operated only by qualified personnel following the guidelines laid out in this instruction manual. Anyone intending to use this equipment should read the complete manual thoroughly. The unit must never be used without the safety lid correctly in position. The unit should not be used if there is any sign of damage to the external tank or lid.

These units comply with the following European directives:

2014/35/EU Low Voltage Directive

2014/30/UE (official Title 2004/108/EC) EMC Electromagnetic Compatibility

By virtue of the following harmonised standards:

BS EN IEC 61010-1: 2010 Safety Testing of Lab Equipment

BS EN IEC 61326-1:2013 EMC Electro Magnetic Compatibility

ROHS DIRECTIVE 2011/65/EU

BS EN 50581:2012 Restriction of Hazardous Substances

Packing List

Each omniPAGE unit includes a tank, wired PAGE module, lid, CSL-CAB cables, S20ICB Cooling pack and the following items:

SKU	Glass Plates	Combs	Caster / Cooling coil	Blotting module
VS10WCBS	VS10WNG - Notched, Pk/2 VS10WPGS1 – Plain with bonded 1mm spacers, Pk/2 VS10W-DP – Dummy Plate	2 of VS20-24-1 1mm thick, 24 sample	VS20DCAST VS20DCASTM - Mat	VS10WBI – Blotting insert 4 of SB10WC – omniBLOT Mini Wide Cassette (including 2 foam pads) SB10WF – Mini Wide Fibre pads Pk/8
VS20CBS	VS20NG - Notched, Pk/2 VS20PGS1 – Plain with bonded 1mm spacers, Pk/2 VS20-DP – Dummy Plate	2 of VS20-24-1 1mm thick, 24 sample	VS20DCAST – Casting base VS20DCASTM – Mat VS20WAVE-CC – Cooling coil	VS20BI – Blotting insert 4 of SB20C – omniBLOT Maxi Cassette (including 2 foam pads) SB20F – Maxi Fibre pads Pk/6

Each omniPAGE Electroblothing unit includes a tank, lid, CSL-CAB cables, VS20ICB Cooling pack and the following items:

SKU	Blotting Module	Cassettes	Fibre pads
SB10W	VS10WBI – Blotting insert	4 of SB10WC – omniBLOT Mini Wide Cassette (including 2 foam pads)	SB10WF – Mini Wide Fibre pads Pk/8
SB20	VS20BI – Blotting insert	4 of SB20C – omniBLOT Maxi Cassette (including 2 foam pads)	SB20F – Maxi Fibre pads Pk/6

Packing List Checked by: _____

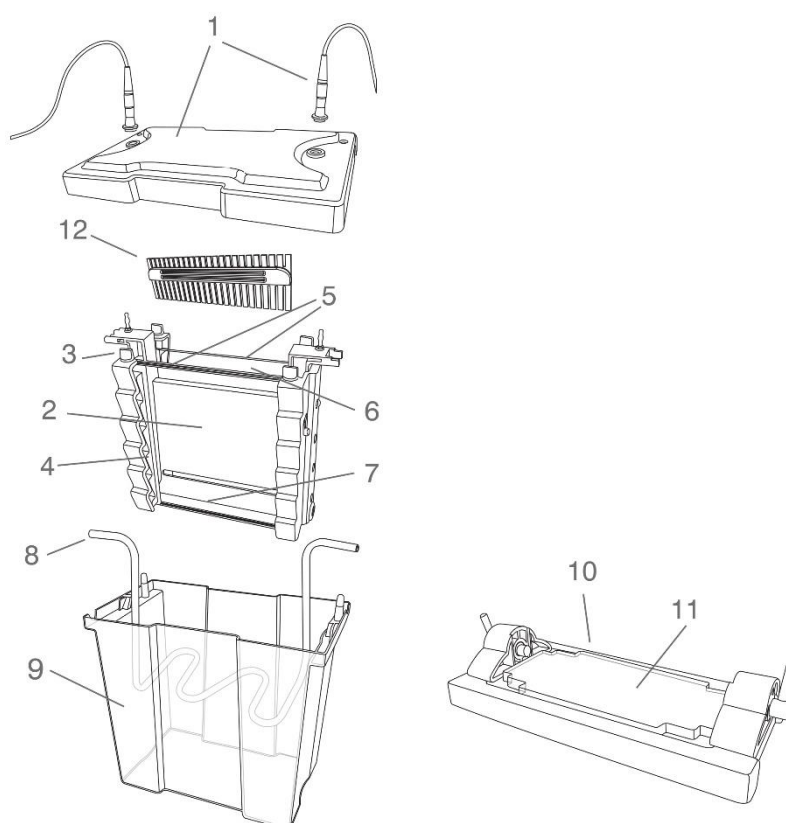
Date: _____

The packing lists should be referred to as soon as the units are received to ensure that all components have been included. The unit should be checked for damage when received.

Thistle Scientific is liable for all missing or damaged parts / accessories within 7 days after customers have received this instrument package. Please contact Thistle Scientific immediately regarding this issue. If no response within such period is received from the customer, Thistle Scientific will no longer be liable for replacement/damaged parts.

Please contact your supplier if there are any problems or missing items.

Specifications



1	Lid and Cables	1500V Rated cables, high impact acrylic lid
2	PAGE Insert	Glass filled Nylon and Acetal construction
3	Clamp Screws	Injection moulded to avoid thread wear
4	Plate Clamps	Injection moulded, provide even pressure across glass plates
5	Glass plates	4mm thick float glass
6	Inner Buffer Chamber	Allows use of separate inner running buffer
7	Electrodes	99.99% pure platinum electrodes
8	Cooling Coil	Can be connected to recirculating chiller
9	Out Buffer Chamber	Injection Moulded High Impact Acrylic
10	Casting Base	Injection Moulded Acetal
11	Casting Mat	Ultra-soft silicone for leak free casting
12	Combs	Lapped Acrylic for high thickness uniformity, also available in polycarbonate
13	Max Voltage	VS20CBS & SB20: 600V DC VS10WCBS & SB10W: 500V

Operating Instructions

Further information (including videos) regarding setting up and running the omniPAGE units can be found at www.thistlescientific.co.uk

Usage Guidance and restrictions

- Maximum altitude 2,000m.
- Temperature range between 4°C and 65°C.
- Maximum relative humidity 80% for temperatures up to 31°C decreasing linearly to 50% relative humidity at 40°C.
- Not for outdoor Use.

This apparatus is rated POLLUTION DEGREE 2 in accordance with IEC 664.

POLLUTION DEGREE 2, states that: "Normally only non-conductive pollution occurs.

Occasionally, however, a temporary conductivity caused by condensation must be expected".

Setting up the omniPAGE Gel Tanks

Fitting Electrode cables

- Note the position of the lid on the unit. This shows the correct polarity and the correct orientation of the cables, black is negative and red positive.
- Remove the lid from the unit. Note if the lid is not removed, fitting the cables may result in un-tightening of the gold plug and damage to the electrode.
- Screw the cables into the tapped holes as fully as possible so that there is no gap between the lid and the leading edge of the cable fitting.
- Refit the lid.

The unit is now ready to be used.

Casting Unit Preparation

Cleaning the Glass Plates

Clean a set of glass plates for each gel first with distilled water and then with 70 % ethanol. One set of glass plates constitutes one notched glass plate and one plain glass plate with bonded spacers.

When using a triple glass plate sandwich, two notched glass plates are required, one set of free spacers and a set of plain glass plates with bonded spacers. The plain glass plate is positioned outermost, then a notched glass plate, free spacers and second notched glass plate. Alternatively, accessory notch glass plates with bonded spacers are available.

Note: All glass plates, gel casting modules, casting base and accessories must be completely dry before the set – up. Wet components are more likely to miss-align and cause leaks.

Gel Cassette Assembly

Assemble the glass plates so that the bottom of the glass plates and the spacers are perfectly aligned.

A triple plate sandwich can be used by combining the standard Notched plate and spacer plate, with an additional notched plate with spacers. For triple plate sandwiches, the free spacers Need to be perfectly aligned which is best performed using a small spacer or comb to push the spacers apart. Notched glass plates with bonded spacers do not need manual alignment.

NOTE: The glass plates with bonded spacers have an arrow in the top of the spacers which are slightly longer than the glass plate to indicate the top.

Casting Stand Assembly



1. Ensure that the clamps are adequately open for the thickness of spacer used. The clamps can be opened by loosening the screws and sliding the clamps.



2. Position the Slab Gel Insert on a flat surface. Insert the glass plates into the Slab Gel Insert between the clamps and the white gasket. The Slab Gel Insert contains clamps which impart even pressure onto the glass plates and allow even screw pressure transfer onto the sealing edge of the glass plate, ensuring complete sealing.



3. Fully tighten the screws, making sure not to wobble the unit.

4. When only one gel is being run, the dummy plate must be used in the second position and fully tightened.



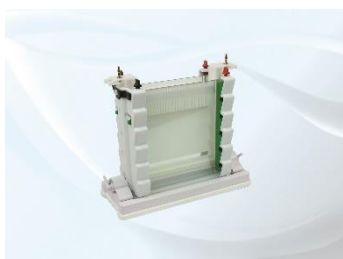
5. Position the Slab Gel Insert in the casting base such that the Cam pins have handles pointing downwards and are located in the insert holes. The top of the module may need to be pushed down very slightly to locate the cam pins.

NOTE: At this stage, check that the bottom edges of the spacers and glass plates are perfectly aligned.



6. With the cam pin handles facing directly downwards, turn the cam pins fully through 135° or until the insert has tightened onto the silicone mat.

NOTE: It is best to turn the cams in opposite directions to each other. Do not overturn as this will cause the glass plates to push upwards and the assembly will be more likely to leak. The unit is now ready for gel preparation and pouring.



7. Pour in the gel solution, insert the combs and allow the wells to polymerise. Gel pouring is best performed using a 25ml or 50ml pipette. If using a stacking gel, don't insert combs, allow gel to polymerise, pour stacking gel and then insert combs.

Casting 4 gels

The thicker green sliding gel clamps are recommended to secure up to 2 gels (i.e. 1 gel either side of the PAGE insert) for gels up to a maximum thickness of 2mm. For 4 gels (i.e. 2 gels either side of the PAGE insert: made using 1 plain glass plate and 1 notched glass plate, both with bonded spacers, and 1 notched plate without spacers), the thinner YELLOW sliding gel clamps must be used.

To convert the unit from a 2- to 4-gel configuration please adhere to the following instructions. One completed, proceed with casting as above.



1. To replace the green sliding gel clamps, begin by unscrewing the colour-coded vertical screw pins. There should be no glass plates within the PAGE insert, in the side(s) being unscrewed. Once the screw pins are unscrewed sufficiently the green clamp should sit in the resting slot as shown.



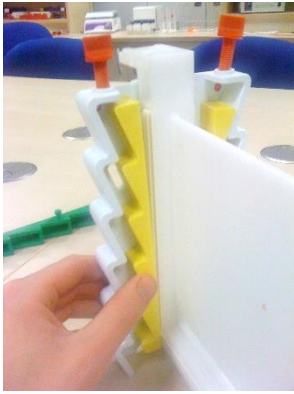
2. Gently push the green sliding clamp horizontally towards the core of PAGE insert until it can move no further.



3. Push the green sliding gel clamp out of the PAGE insert by gently pressing the protruding pin as shown.



4. Once the green sliding gel clamp is removed the PAGE insert is ready to accept the thinner yellow sliding gel clamps for 2-gel sandwiches, either side of the PAGE insert, to convert the WAVE to a 4-gel configuration.



5. Insert the yellow sliding gel clamp into the hole closest to the core of the PAGE insert. Once inserted gently withdraw the sliding clamp outwards away from the core of the PAGE insert as shown.



6. Withdraw the sliding gel clamps so that they sit suspended in the resting slots as shown. Repeat steps 1-6 to replace the remaining green sliding gel clamps. Once complete the PAGE insert is ready for 4-gel assembly.

Gel Preparation

Care should be taken when selecting the pore size of the gel to be used. The pore size or % of gel determines the resolving ability given different sizes of protein.

Gel percentage for various protein sizes are shown below:

Acrylamide Percentage	Separating Resolution
5 %	60 - 220 KD
7.5 %	30 - 120 KD
10 %	20 - 75 KD
12%	17 – 65 KD
15 %	15 -45 KD
17.5%	12 – 30 KD

Gel Volumes

For omniPAGE Maxi

Number of Gels	Volume for 1mm Thick Gel
Single	35 ml
Double	70 ml
4 Gels (2 x Triple plate)	140 ml

For omniPAGE Mini Wide

Number of Gels	Gel Thickness (mm)	Volume (mm)
Single	0.5	8.75
	1	17.5
	1.5	26.3
	2	35
Double	0.5	17.5
	1	35
	1.5	52.5
	2	70
4 Gels (2 x Triple plate)	0.5	35
	1	70
	1.5	105
	2	140

Gel Mixture

Prepare gel solutions as per tables below. These give the volumes of solutions from the standard stock solutions. These should be gently mixed avoiding generation of bubbles which will inhibit polymerization by removing free radicals. The following values are for a 1 mm gel for the omniPAGE Maxi, adjust volumes accordingly for other gel sizes.

Resolving Gel:

Solution	5 %	7.5%	10 %	12%	15 %	17.5%
Distilled Water	41ml	35.25ml	29.6ml	24.7ml	17.6ml	11.7ml
30 % Stock Acrylamide Solution	11.7ml	17.6ml	23.5ml	28.2ml	35.25ml	41.1ml
4 X Resolving Tris Solution	17.6ml	17.6ml	17.6ml	17.6ml	17.6ml	17.6ml

Before pouring, add and mix 700 µl of 10% ammonium persulfate and 70 µl of TEMED, these are the polymerising agents and will initiate polymerisation of the gel, so must be added only immediately before pouring.

Stacking Gel

Solution	VS20WAVE	VS10W
Distilled Water	16.8ml	8.4ml
30 % Stock Acrylamide Solution	2.6ml	1.3ml
4 X Stacking Gel Tris Solution	6.4ml	3.2ml

Before pouring, add and mix 178 µl of 10% ammonium persulfate and 17.8 µl of TEMED for an omniPAGE Maxi gel or 134 µl of ammonium persulfate and 13.4 µl of TEMED for an omniPAGE Mini Wide gel. These are the polymerising agents and will initiate polymerisation of the gel, so must be added only immediately before pouring.

Gel Pouring

With Stacking Layer

1. Insert the comb into the glass plates and mark a point on the glass plates 1cm below where the comb teeth finish. This indicates where to add the resolving gel to.
2. Add polymerising agents to the gel solution and mix well but avoid generating air bubbles.

3. Fill the glass plates again avoiding generating any air bubbles. Filling must be performed quickly before the gel becomes too viscous.
4. Overlay the gel carefully with 1 ml of Isobutanol, Isopropanol or distilled water. When using distilled water extra care must be taken to ensure there is no mixing with the gel solution.
5. Let the resolving gel polymerize. Usually this takes around 15 minutes, but this can vary due to the freshness of the reagents used. If polymerization takes a lot longer than this, use fresher stock solutions or add more APS and TEMED.
6. Carefully mix the stacking gel solution, avoiding generating air bubbles.
7. Pour off the overlay liquid from the resolving gel and rinse the gel with distilled water.
8. Add polymerising agents to the stacking gel and pour over the resolving gel.
9. Carefully insert the comb making sure that no air bubbles get trapped under the ends of the comb teeth as these will inhibit sample progression.
10. Allow the stacking gel to polymerize for 30 minutes.

With Stacking Layer

1. Add polymerising agents to the gel solution and mix well but avoid generating air bubbles.
2. Fill the glass plates again avoiding generating any air bubbles. Filling must be performed quickly before the gel becomes too viscous.
3. Carefully insert the comb making sure that no air bubbles get trapped under the ends of the comb teeth as these will inhibit sample progression.
4. Let the resolving gel polymerize. Usually this takes around 15 minutes, but this can vary due to the freshness of the reagents used. If polymerization takes a lot longer than this, use fresher stock solutions or add more APS and TEMED.

Sample preparation and Loading

Denatured sample preparation

The instructions given below are for denatured samples. For Native samples, please consult a laboratory handbook.

1. Prepare the protein samples for loading. The volume of sample depends on the capacity of the wells (See Comb specifications page 28).

2. Using a 0.5 ml micro-centrifuge tube or other convenient receptacle, combine the protein sample and 4 X sample buffer. It is always advisable to use protein markers in one of the end lanes to indicate sizes of bands. These should be prepared according to the manufacturer's instructions.
3. Heat the samples in a water bath or heating block for 2 minutes to denature the samples.
4. Centrifuge the samples in a micro-centrifuge for 20 seconds at 12,000 rpm. The protein samples are now ready to load.

Loading the samples

1. If desired, fit the cooling pack(s) into the end of the tank. These should be pre-frozen and fitted with the longest side positioned sideways with the end(s) of the tank and pressed into the recess. Or these can be fitted down the front of the tank.

NEVER FIT THESE UNDERNEATH THE MODULE IN THE BOTTOM OF THE TANK AS THIS WILL PREVENT THE FLOW OF CURRENT THROUGH THE GEL AND CAUSE SLOW RUNS AND OVER-HEATING.

2. Note one pack is supplied as standard. Additional packs can be purchased.
3. Transfer the Inner gel module containing cast gels into the main tank in the correct orientation as indicated - +ve on the module aligned with +ve on the tank, -ve on the module aligned with -ve on the tank.
4. Fill the outer tank with 1X reservoir buffer.
5. Load the samples into the wells using a pipette tip taking care not to damage the wells or induce any air bubbles.
6. Fill any unused wells with 1X sample buffer.
7. It is a good idea to note the orientation and order the samples were loaded in. This can be done by noting which samples were loaded adjacent to each electrode.

Buffer Volumes

Buffer Volume	VS20WAVE	VS10W
Minimum – Inner tank is filled to above the wells. Outer Tank is filled to just flood the bottom of the glass plates. Cooling potential is at a minimum which may affect resolution.	Total Inner Buffer Chamber: 640ml Outer Tank: 1L	Total Inner Buffer Chamber: 300ml Outer Tank: 500ml
Maximum – Inner tank is filled to above the wells. Outer Tank is filled to the maximum fill line. Cooling is high offering good resolution of samples. This may be further enhanced by using the cooling coil	Total Inner Buffer Chamber: 640ml Outer Tank for 2 gels: 5.4L Outer Tank for 4 gels: 4.8L	Total Inner Buffer Chamber: 300ml Outer Tank for 2 gels: 2.8L Outer Tank for 4 gels: 2.3L

Gel Running

1. Fit the lid and connect to a power supply.
2. Consult the below table for details on recommended power supply voltage settings.
3. Turn the power supply off when the loading dye reaches the bottom of the gel, sooner if your proteins are below 4Kd in size.
4. Remove the gel running module, first emptying the inner buffer into the main tank. Buffer can be re-used but this may affect run quality if continued.
5. Unscrew the glass plates and gently pry apart the glass plates. The gel will usually stick to one of the plates and can be removed by first soaking in buffer and then gently lifting with a spatula.
6. The gel is now ready to be stained with Coomassie or silver stain or the proteins in the gel can be transferred to a membrane by electroblotting for specific band identification and further analysis.

Recommended Voltages for 1mm thick, 12% gels.	VS20WAVE	VS10WD
2-4 gels	Constant Voltage 250V 6 Hours	Constant Voltage 125V 1 Hour

We recommend that for any new sample, a time course experiment is performed to obtain the optimal running conditions.

Connecting a recirculating chiller

Vertical PAGE may sometimes result in high currents. High current may cause the build-up of heat which can affect sample migration and gel resolution as a result. To counteract the adverse effects of heat generation, the manufacturer recommends using an effective chiller unit, such as the CSL-CHILLER, pre-set to 4°C.

1. For active temperature regulation attach two short lengths of hose from the inlet and outlet ports of the chiller unit to the respective outlet and inlet connectors of the cooling coil sitting within the WAVE outer tank.
2. Ensure that the flow rate of the chiller unit is set at an acceptable value by first testing with the cooling coil outside of the electrophoresis tank.

The unit is now ready to be used with active cooling.

Solutions (For SDS-PAGE)

Stock 30% Acrylamide Gel Solution:

30.0 g acrylamide

0.8 g methylene bisacrylamide

Distilled Water to 100ml

Stock 4 X Resolving Gel Tris (1.5 M Tris·HCl pH8.8, 0.4 % SDS)

To 110ml Distilled Water add 36.4 g of Tris base

Add 8ml of 10 % SDS

Adjust pH to 8.8 with 1N HCl

Adjust the final volume to 200ml with Distilled Water.

Stock 4 X Stacking Tris (0.5 M Tris·HCL pH6.8, 0.4 % SDS)

To 110ml Distilled Water add 12.12 g of Tris base

Add 8ml of 10 % SDS

Adjust pH to 6.8 with 1N HCl

Add Distilled Water to a final volume of 200ml

Stock 4 X Tris-glycine tank buffer - SDS

36 g Tris base

172.8 g glycine

Distilled Water to 3 L

1 x Tris-glycine tank buffer - SDS

750ml of 4 X Tris-glycine reservoir buffer - SDS

30ml of 10 % SDS

Distilled Water to 3L

10 % AP (ammonium persulphate solution)

0.1 g ammonium persulphate

1ml Distilled Water

TEMED

Stock 4 X Sample Buffer

4ml glycerol

2ml 2-mercaptoethanol

1.2 g SDS

5ml 4 X Stacking Tris

0.03 g Bromophenol blue

Aliquot into 1.5ml micro centrifuge tubes. Store at -20°C.

References

1. Sambrook, Fritsch, and Maniatis, Molecular Cloning A Laboratory Manual, Second Edition, Cold Spring Harbor Laboratory Press, 1989.
2. Current Protocols in Molecular Biology, Greene Publishing Associates and Wiley-Interscience, 1989.

Protein Blotting using the omniPAGE Electroblotter

Setting up the blot sandwich

N.B Remember to always wear gloves when dealing with proteins to avoid contaminations!

1. Following electrophoresis, remove the glass plate and transfer the gel into Transfer buffer.
2. Equilibrate the gel between 10-30min at room temperature on a shaker according to specific protocols.
3. Cut the membrane of the same size of the gel and equilibrate in Transfer buffer as well.

N.B it is best to assemble the blotting sandwich in some transfer buffer to avoid the membrane to dry out.

Each blot sandwich should be set up as follows:

- a. Cassette clamp -ve (black) side placed in a tray or other suitable surface
 - b. Pre-soaked fibre pad. Note two can be used with thin gels.
 - c. Two pieces of thick filter paper, about 2 – 3 mm thick, pre-soaked in buffer.
 - d. Gel.
 - e. Transfer membrane. Usually this requires pre-soaking but consult the manufacturers instructions for the type of membrane you are using. This should be smoothed so that no air bubbles have been trapped.
 - f. Two pieces of thick filter paper, about 2 – 3 mm thick, pre-soaked in buffer.
 - g. Pre-soaked fibre pad. Note two can be used with thin gels.
 - h. Cassette clamp +ve (red) side slotted into the groove in the bottom of the black cassette.
2. Close the hinge carefully so as to not disturb the sandwich.
 3. Fill the tank with buffer solution up to the maximum fill line indicated on the side of each unit. See page xx for recommended buffer solutions. Improved transfer can usually be obtained by using chilled buffer.

Blot Run Conditions

1. Insert the cassettes into the slots in the module with the black side of each adjacent to the negative electrode. It is a good idea to note the orientation and order the blot sandwiches were loaded in. This can be done by noting which samples were loaded adjacent to each electrode.
2. Use of a magnetic stirring bar and plate is recommended to mix the buffer to give consistency of transfer. A 4mm diameter stirring bar should be placed underneath the module, in the centre of the tank. The Cooling pack provided, pre-frozen, can be inserted at the side or front of the tank for extended blots. Additional cooling packs can be purchased as accessories to further aid cooling.
3. Insert the module, fit the lid and connect to a power supply.
4. Consult Suggested Running Condition for recommended power supply voltage settings and blot times. Please note voltages and current will vary according to the number of cassettes, type and temperature of buffer and thickness and percentage of gel. This will also affect quality of transfer so time course of transfer should be performed for your particular samples and conditions.
5. When the blot time is completed, turn the power supply off.
6. Remove the cassettes from the main tank. Buffer can be re-used but this may affect run quality if continued.
7. Lift the hinge of each cassette and gently pry apart the blot sandwich and remove the membrane from the gel.
8. The membrane is now ready to be probed.

Suggested Running Conditions (please note that these are just guideline)

Duration of Blot	VS10W / BS10W	VS20 / SB20
High Intensity	100 V 380–500 mA 60 min	100–200 V 300–800 mA 30 min–4 hr
Overnight	10 V 30–40 mA 16 hr	30 V 100 mA 16 hr

N.B. If the voltage is held constant throughout a transfer, the current in most transfer systems increases as the resistance drops due to heating. Therefore, the overall power increases during transfer, and more heating occurs. Despite the increased risk of heating, a constant voltage ensures that field strength remains constant, providing the most efficient transfer possible for tank blotting methods. Use of the cooling elements available with the various tank blotting systems should prevent problems with heating.

Buffer Solutions (For Blotting)

Towbin Buffer

25mM Tris,
192mM glycine,
20% methanol pH8.3,

Towbin Buffer SDS

25mM Tris
192mM glycine
20% methanol pH8.3
0.05-0.1% (w/v) SDS

Bjerrum and Schafer-Nielsen Buffer

48mM Tris

39mM glycine

20% methanol pH9.2

Dunn Buffer

10mM NaHCO₃

3mM NaCO₃

20% methanol pH9.9

N.B. Do not adjust the pH when making these buffers as this will cause blot over-heating.

The pH will vary according to the freshness of the reagents used.

Troubleshooting

SDS-PAGE electrophoresis		
Problem: Sample Preparation	Cause	Solution
Laemmli sample buffer turns yellow	Sample buffer too acidic	Add Tris base until buffer turns blue again.
Sample very viscous	High DNA or carbohydrate content	Fragment DNA with ultrasonic waves during cell lysis and protein solubilization. Add endonucleases (for each benzonases). Precipitate protein with TCA/acetone to diminish carbohydrate content.
Problem: Gel casting and sample loading	Cause	Solution
Poor well formation	Incorrect catalyst used Monomer solution not degassed (oxygen inhibits polymerization)	Prepare Fresh catalyst solution. Increase catalyst concentration of stacking gel to 0.06% APS and 0.12% TEMED. Degas monomer solution immediately prior to casting stacking gel.
Webbing; excess acrylamide behind the comb	Incorrect catalyst concentration	Prepare fresh catalyst solution. Increase catalyst concentration of stacking gel to 0.06% APS and 0.12% TEMED.
Gel does not polymerize	Too little or too much APS or TEMED Failure to degas Temperature too low Poor quality acrylamide or bis Old APS	Use 0.05% APS and 0.05% TEMED. Degas monomer solutions 10-15min. Cast at room temperature, warming glass plates if necessary. Use electrophoresis-grade reagents Prepare fresh APS.
Swirls in the gel	Excess catalysts; polymerization time < 10min Gel inhibition; polymerization time > 2hr	Reduce APS and TEMED by 25% each. Increase APS and TEMED by 50%; degas.
Gel feels soft	Low %T Poor quality acrylamide or bis Too little cross-linker	Use different %T. Use electrophoresis-grade reagents. Use correct %C.
Gel turns white	Bis concentration too high	Check solutions or weights.
Gel brittle	Cross-linker too high	Use correct % cross-linker
Sample floats out of the well	Sample is not dense enough Pipetting, loading error	Induce 10% glycerol in sample to make it denser than surrounding buffer. Slowly pipet sample into well. Do not squirt sample quickly into well as it may bounce off bottom or sides and flow into next well. Do not pipet tip from well before last of sample has left the tip.
Problem: Electrophoresis	Cause	Solution

Current zero or less than expected and samples do not migrate into gel	Tape at the bottom of precast gel cassette not removed Insufficient buffer in inner buffer chamber Insufficient buffer in outer buffer chamber Electrical disconnection	Remove tape. Fill buffer chamber with running buffer. Fill inner and outer buffer chambers to ensure wells are completely covered. Check electrodes and connections.
Gels run faster than expected	Running buffer too concentrated and gel temperature too high; incorrect running buffer concentration or type used Running or reservoir buffer too dilute Voltage too high	Check buffer composition and type. Check buffer protocol and concentrate if necessary. Decrease voltage by 25-50%.
Gels run slower than expected	Incorrect running buffer composition or type Excessive salt in sample	Check buffer composition and type. Desalt sample.
Problem: Total Protein Staining	Cause	Solution
Bands not visible	No protein in gel Imaging system malfunctioning Incorrect imaging parameters were used	Stain with another method to confirm there is protein. Check instrument manual for troubleshooting or contact imaging instrument manufacturer. Check Instrument manual.
Poor staining sensitivity	Dirty staining trays Insufficient stain volume Insufficient staining time Reuse of staining solution	Clean staining trays and other equipment with laboratory glassware cleaner. Follow recommendations for stain volume (appropriate to gel size). Increase staining time. Repeat staining protocol with fresh staining solution.
High or uneven background staining	Staining trays or equipment dirty Too much time in staining solution Reagent impurities	Clean staining trays and other equipment with laboratory glassware cleaner. Restrict duration of incubation in staining solutions as recommended in protocol. Wash gel in water or retrospective destaining solution for >30min. Use high-purity water and reagents for staining.
Speckles or blotches in gel image	Particulate material from reagents, staining tray, dust or gloves	Clean staining trays thoroughly. Decrease time that gels and staining solution are exposed to open air. Use dust-free gloves and handle gels only by edges.
Uneven staining	Insufficient shaking during staining	Agitate gel during staining.
Gel shrinkage	Gel dehydrated	Transfer gel to water for rehydration.
Problem: Evaluation of Separation	Cause	Solution
Diffuse or broad bands	Poor quality acrylamide or bis-acrylamide incomplete polymerization Old SDS or sample buffer Gel temperature too high	Use electrophoresis-grade reagents. Check polymerization conditions. Prepare fresh solutions. Use external cooling during run or run out a lower voltage.

Bands 'smile' across gel, band pattern curves upward at both sides of gel	Excess heating of gel; centre of gel runs hotter than either end Power conditions excessive Insufficient buffer	Check buffer composition; buffer not mixed well, or buffer in upper chamber too concentrated. Prepare new buffer, ensuring thoroughly mixing, especially when diluting 5x or 10x stock. Do not exceed recommended running conditions. Decrease power setting from 200V to 150V or fill lower chamber to within 1 cm of top of short plate. Fill inner and outer buffer chambers to ensure that wells are completely covered.
Smiling or frowning bands with gel lane	Overloaded proteins Sample preparation/ buffer issues Incorrect running conditions	Load less protein. Minimize salts, detergents and solvents in sample preparation and sample buffers. Use correct voltage.
Skewed or distorted bands, lateral band spreading	Excess salt in samples Ionic strength of sample lower than that of gel Insufficient sample buffer or wrong formulation Diffusion prior to turning on current Diffusion during migration through stacking gel Uneven gel interface	Remove salts, from sample by dialysis or de-salting column prior to sample preparation. Use same buffer in samples as in gel. Check buffer composition and dilution instructions. Minimize time between sample application and power start-up. Increase %T of stacking gel to 4.5% or 5%T. Increase current by 25% during stacking. Decrease polymerization rate. Overlay gels carefully. Rinse wells after removing comb to remove residual acrylamide.
Vertical streaking	Overloaded samples Sample precipitation	Dilute sample. Selectively remove predominant protein in sample (fractionate). Reduce voltage by 25% to minimize streaking. Centrifuge samples to remove particulate prior to sample loading. Dilute sample in sample buffer.
Fuzzy or spurious artefactual bands	Concentration of reducing agent too low	Use 5% BME or 1% DTT.
Protein bands do not migrate down as expected	Bands of interest may be neutral or positively charged in buffer used; pH of bands must be -2pH units more negative than pH of running buffer	Use SDS-PAGE or a different buffer system in native PAGE or IEF.

Blotting

Poor protein transfer	Transfer apparatus assembled incorrectly and proteins moving in the wrong direction. - Gel/membrane sandwich may be assembled in the wrong order, or cassette inserted in wrong orientation. Check polarity. - Air pockets not removed while assembling the blotting sandwich
	Western detection system not working or not sensitive enough Include proper positive or negative control antigen. Consult kit manual.

	• Use protein markers with coloured reference bands during PAGE. • Stain gel with Coomassie, or stain membrane with Ponceau S.
	Transfer time too short – increase transfer time
	Power setting too low • Increase Voltage • Buffer may be prepared improperly – prepare new buffer and increase voltage.
	Charge-to-mass ratio incorrect for native transfers. • Proteins close to isoelectric point (pI). Change buffer pH so that it is at least 2 pH unit higher or lower than pI of protein of interest.
	Defective or inappropriate power supply used. • Check fuse of power supply. Ensure max. current output of power supply is at least 2000mA.
	Excessive methanol restricting transfer. • Reduce methanol concentration to maximize protein transfer from gel, but without reducing concentration to the extent that it prevents binding to nitrocellulose. Alternatively reduce methanol concentration and switch to PVDF.
Protein precipitating in gel	• Use SDS in transfer buffer (SDS can increase transfer efficiency, but it can also reduce nitrocellulose binding affinity and affect protein-antibody reactivity). • Remove alcohol from transfer buffer.
Swirls or missing bands; diffuse transfers	Poor gel-membrane contact. Air bubbles or excess buffer remain between membrane and gel. • Carefully remove air bubbles between gel and membrane using a rolling pin • Use more, or thicker, filter paper in gel-membrane sandwich • Replace the fibre pads, as they degrade and remain permanently compressed over time.
	Problem with gel electrophoresis. • Poor gel polymerization, inappropriate running conditions, buffer contamination, excessive sample application all contribute to poor quality gels and transfers.
Gel cassette pattern transferred to blot	Contaminated fibre pads • Replace fibre pads or clean thoroughly. Contaminated transfer buffer • Replace buffer solutions.
Poor binding to membrane - nitrocellulose	Excessive methanol restricting transfer. • Ensure methanol concentration does not exceed 20% (v/v).
	Proteins may be transferring through nitrocellulose. • Use PVDF or smaller pore size (0.2µm) nitrocellulose.

	Overlay an extra piece of nitrocellulose over membrane to determine if proteins are migrating through the membrane directly in contact with the gel.
	Proteins <15kDa have reduced binding to 0.45µm nitrocellulose or may be washed from membrane during assays. - Use PVDF or nylon membrane, which have higher binding capacities. - Use Tween-20 detergent in the wash and antibody incubation steps. Reduce or eliminate the more stringent washing steps.
	SDS in transfer buffer reducing binding efficiency Reduce or eliminate SDS concentration
	Membrane incompletely wet - White spots indicate dry areas where protein will not bind. - If soaking does not occur immediately following immersion in transfer buffer, heat distilled water to just below boiling point and soak membrane until entirely wet.
Poor binding to membrane PVDF	Membrane is not completely wet Because of hydrophobicity of PVDF, the membrane must be soaked entirely in methanol before equilibration in aqueous buffer
	Proteins might be transferring through the membrane - Decrease voltage if transferring under high intensity conditions - Overlay an extra piece of PVDF over membrane to determine if proteins are migrating through the membrane directly in contact with the gel.
	Membrane might have dried during handling Fully wet membranes have a grey translucent appearance. White spots will form on the surface if the membrane has been allowed to dry. As proteins will not bind to dry spots, re-soak the membrane in methanol and re-equilibrate in transfer buffer
	SDS in transfer buffer reducing binding efficiency Reduce or eliminate SDS concentration
Power is too high	Always check current at the start of the run, for the current might be too high for a given voltage setting. Improper buffer preparation can also result in high conductivity and excessive power generation. The current setting should not be allowed to exceed 2000mA.
Immune-specific detection	Overall high background - Reduce antibody / protein sample concentration Too low background - Increase antibody concentration / protein sample concentration Consult manual included with antibody detection kit
Total protein detection	Consult stain or detection kit manual.

Care and Maintenance

Cleaning Vertical Units

Units are best cleaned using warm water and a mild detergent. **Water at temperatures above 60°C can cause damage to the unit and components.**

The tank should be thoroughly rinsed with warm water or distilled water to prevent build-up of salts, but care should be taken not to damage the enclosed electrode, and vigorous cleaning is not necessary or advised.

Air drying is preferably before use.

The units should only be cleaned with the following:

Warm water with a mild concentration of soap or other mild detergent.

Compatible detergents include dishwashing liquid, Hexane and Aliphatic hydrocarbons

The units should not be left in detergents for more than 30 minutes.

The units should never come into contact with the following cleaning agents, these will cause irreversible and accumulative damage:

Acetone, Phenol, Chloroform, Carbon tetrachloride, Methanol, Ethanol, Isopropyl alcohol, Alkalies.

RNase Decontamination

This can be performed using the following protocol:

Clean the units with a mild detergent as described above.

Wash with 3% hydrogen peroxide (H₂O₂) for 10 minutes.

Rinsed with 0.1% DEPC- (diethyl pyro carbonate) treated distilled water,

Caution: DEPC is a suspected carcinogen. Always take the necessary precautions when using.

RNaseZAP™ (Ambion) can also be used. Please consult the instructions for use with acrylic gel tanks.

Ordering information

Catalogue No.	Product Description
VS10WD	omniPAGE Mini Wide, 20 x 10cm Dual, 2 sets of Glass Plates with 1mm thick bonded Spacers, 2 x 24 sample, 1mm thick combs, cooling pack
VS10WDSYS	omniPAGE Mini Wide, 20 x 10cm Dual, 2 sets of Glass Plates with 1mm thick bonded Spacers, 2 x 24 sample, 1mm thick combs, cooling pack including caster
VS10WDSYS-CU	omniPAGE Mini Wide, 20 x 10cm Dual, 2 sets of Glass Plates, 1mm thick bonded Spacers, 2 x 24 sample, 1mm thick combs including caster, External casting upstand
VS10WEXCASTER	VS10W External Casting Stand - No Casting Base
VS20CAST	20 x 10cm Casting Base
VS20DCASTM	Replacement Silicone Mat for 20 x 10cm Casting Base
VS10WDIRM	Inner Running Module
VS20-x –LG	Loading guides for omniPAGE mini combs, x = comb well number
VS10WNG	20 x 10cm Notched Glass Plates 4mm thick (pk/2)
VS10WPG	20 x 10cm Plain Glass Plates 4mm thick (pk/2)
VS10WNGS0.75	20 x 10cm Notched Glass Plates with 0.75mm Bonded Spacers (pk/2)
VS10WPGS0.75	20 x 10cm Plain Glass Plates with 0.75mm Bonded Spacers (pk/2)
VS10WNGS1	20 x 10cm Notched Glass Plates with 1mm Bonded Spacers (pk/2)
VS10WPGS1	20 x 10cm Plain Glass Plates with 1mm Bonded Spacers (pk/2)
VS10WPGS1.5	20 x 10cm Plain Glass Plates with 1.5mm Bonded Spacers (pk/2)
VS10WPGS2	20 x 10cm Plain Glass Plates with 2mm Bonded Spacers (pk/2)
VS10WDP	Dummy Plate, 20 x 10cm
RPW-0.2	Replacement Platinum Wire - 0.2mm, 50cm
VS20ICB	Maxi Cooling Pack
VS20WAVESYS	VS20WAVE Maxi, 20 x 20cm Dual with Glass Plates with bonded 1mm thick spacers, 2x 24 sample combs, cooling coil, dummy plate and Casting Base
VS20WAVESYS-CU	VS20WAVE Maxi, 20 x 20cm Dual, 2 sets of Glass Plates, 1mm thick bonded Spacers, 2 x 24 sample, 1mm thick combs, cooling coil, dummy plate; includes caster and External casting upstand
VS20WAVE-EC	VS20WAVE External Casting Stand - No Casting Base
VS20DCAST	20 x 20cm Dual Casting Base
VS20DCASTM	Replacement Silicone Mat for 20 x 20cm Casting Base
VS20WAVEDIRM	PAGE insert
VS20WAVE-CC	Detachable Cooling Coil
VS20-x –LG	Loading guides for omniPAGE maxi combs, x = comb well number

VS20NG	20 x 20cm Notched Glass Plates 4mm thick (pk/2)
VS20PG	20 x 20cm Plain Glass Plates 4mm thick (pk/2)
VS20NGS0.75	20 x 20cm Notched Glass Plates with 0.75mm Bonded Spacers (pk/2)
VS20PGS0.75	20 x 20cm Plain Glass Plates with 0.75mm Bonded Spacers (pk/2)
VS20NGS1	20 x 20cm Notched Glass Plates with 1mm Bonded Spacers (pk/2)
VS20PGS1	20 x 20cm Plain Glass Plates with 1mm Bonded Spacers (pk/2)
VS20PGS1.5	20 x 20cm Plain Glass Plates with 1.5mm Bonded Spacers (pk/2)
VS20PGS2	20 x 20cm Plain Glass Plates with 2mm Bonded Spacers (pk/2)
VS20DP	Dummy Plate, 20 x 20cm
VS20S0.75	20cm Spacers - 0.75mm (pk/2)
VS20S1	20cm Spacers - 1mm thick (pk/2)
VS20S1.5	20cm Spacers - 1.5mm thick (pk/2)
VS20S2	20cm Spacers - 2mm thick (pk/2)
VS20WAVE-IEFKIT	IEF Conversion for 18cm IPG strips and tube gels, includes: 1 set of plain glass plates with bonded spacers, 0.6x20cm (WxH); and 2x 2-D combs with one 3.5mm marker lane and one 18cm preparatory well
RPW-0.2100	Replacement Platinum Wire - 0.2mm, 100cm

Comb options

Catalogue No.	Thickness	Sample Number	Sample Volume µl per well
VS20-1-0.75	0.75	1	1100
VS20-5-0.75	0.75	5	160
VS20-10-0.75	0.75	10	80
VS20-18MC-0.75	0.75	18	40
VS20-24-0.75	0.75	24	30
VS20-30-0.75	0.75	30	25
VS20-36-0.75	0.75	36	20
VS20-48-0.75	0.75	48	15
VS20-1-1	1	1	1500
VS20-5-1	1	5	200
VS20-10-1	1	10	100
VS20-18MC-1	1	18	50
VS20-24-1	1	24	40
VS20-30-1	1	30	35
VS20-36-1	1	36	25

VS20-48-1	1	48	20
VS20-1-1.5	1.5	1	2200
VS20-5-1.5	1.5	5	320
VS20-10-1.5	1.5	10	160
VS20-18MC-1.5	1.5	18	80
VS20-24-1.5	1.5	24	60
VS20-30-1.5	1.5	30	50
VS20-36-1.5	1.5	36	40
VS20-48-1.5	1.5	48	30
VS20-1-2	2	1	3000
VS20-5-2	2	5	400
VS20-10-2	2	10	200
VS20-18MC-2	2	18	100
VS20-24-2	2	24	80
VS20-30-2	2	30	70
VS20-36-2	2	36	50
VS20-48-2	2	48	40

MC denotes multichannel pipette spacing

Related Products

Catalogue No.	Product description
CSL-PPL	CSL Pink Plus Prestained Protein Ladder, 10-175kDa, with 10, 40 & 90kDa reference bands, 1x 500µL vial.
CSL-BBL	CSL BLUE Wide Range Prestained Protein Ladder, 10-245kDa, with 25 & 75kDa reference bands, 1x 500µL vial.
CSL-TGSDSP	Powdered Tris-Glycine-SDS Running buffer - 10 Pouches (10 litres/pk)
CSL-TGP	Powdered Tris-Glycine Running buffer - 10 Pouches (10 litres/pk)
CSL-TTSDSP	Powdered Tris-TRICINE-SDS Running buffer - 10 Pouches (10 litres/pk)
CSL-MSDSP	Powdered MOPS-SDS buffer Running buffer - 10 Pouches (10 litres/pk)
TG10X1L	Cleaver Buffer Tris-Glycine 10 x 1litre
TG10x5L	Cleaver Buffer Tris-Glycine 10 x 5litre
TG-SDS10X1L	Cleaver Buffer Tris-Glycine SDS 10 x 1litre
TG-SDS10X5L	Cleaver Buffer Tris-Glycine SDS 10 x 5litre
CSL-GELX4	4mm x 1mm, Gel Cutting Tips, 250/ bag
CSL-GELX4RACK	4mm x 1mm, Gel Cutting Tips, 5 racks of 48

CSL-GELX6.5	6.5mm x 1mm, Gel Cutting Tips, 250/ bag
CSL-GELX6.5RACK	6.5mm x 1mm, Gel Cutting Tips, 5 racks of 48
POWERPRO300	Power Supply MIDI 300V 700mA 150W - 110/230V
POWERPRO500	Power Supply MAXI 500V 800mA 200W - 110/230V
VS10BI	omniBlot Mini Insert - including 4 cassettes, 16 foam pads
VS10DCI	omniPAGE Mini Tube Gel Insert - including 10 glass tubes
OMNIDOCIPROSAFE	OMNIDOC-i plus Blue LED Epi-illumination Module (OMNIDOC-BL), and 520, 560 & 580nm filters (OMNIDOC-SYBR, -AF560 & -AF580); and White Light Table (OMNIDOC-WLT)
CVS10CBS	Complete system for Mini Vertical Electrophoresis & Blotting including Vertical unit, Blotting insert & accessories. CLAMP VERSION.
CV20	Cleaver Pipette - Volume; 2 - 20ul
CSLVORTEX	Cleaver Vortex Mixer with general purpose head, 230V
CSLQSPIN	Mini Centrifuge complete with 1.5/2.0 ml rotor, strip tube rotor, 0.5- and 0.4-ml adapters, 230V, Purple lid
TCDB-01	The Cube Dry Bath Incubator (one block unit); without block 220V
CSL-UVCAB	UV sterilisation cabinet with timer, four UV lights and white lights, no Tray - 230V
SD20	Semi Dry Maxi, 20 x 20cm System
CVS10D	omniPAGE Mini, 10 x 10cm Dual, 2 sets of Glass Plates, 1mm thick bonded Spacers, 2 x 12 sample, 1mm thick combs. CLAMP VERSION
CVS10DSYS	omniPAGE Mini, 10 x 10cm Dual, 2 sets of Glass Plates, 1mm thick bonded Spacers, 2 x 12 sample, 1mm thick combs including caster. CLAMP VERSION
CVS10PRE	omniPAGE Mini, 10 x 10cm Dual. No accessories. CLAMP VERSION
CVS10DSYS-CU	omniPAGE Mini, 10 x 10cm Dual, 2 sets of Glass Plates, 1mm thick bonded Spacers, 2 x 12 sample, 1mm thick combs including caster. CLAMP VERSION, External casting upstand

Warranty

The Thistle Scientific Ltd. (TSL) omniPAGE Horizontal Electrophoresis units have a warranty against manufacturing and material faults of thirty-six months from date of customer receipt.

If any defects occur during this warranty period, TSL will repair or replace the defective parts free of charge.

This warranty does not cover defects occurring by accident or misuse or defects caused by improper operation.

Units where repair or modification has been performed by anyone other than TSL or an appointed distributor or representative are no longer under warranty from the time the unit was modified.

Units which have accessories or repaired parts not supplied by TSL or its associated distributors have invalidated warranty.

TSL cannot repair or replace free of charge units where improper solutions or chemicals have been used. For a list of these please see the Care and Maintenance subsection.

This warranty does not apply to parts listed below:

1. Platinum Electrode Wires.

If a problem does occur, then please contact your supplier or Thistle Scientific Ltd:

Thistle Scientific Ltd.

Unit 41, Somers Road Industrial Estate

Rugby, Warwickshire, CV22 7DH

Tel: +44 (0)1788 565300

Email: enquiries@thistlescientific.co.uk