One-Dimensional SDS Electrophoresis

1D SDS PAGE kits contains ready-to-use SDS polyacrylamide gels, running buffers, paper wicks, and sample diluters are available from **gel**company. The gels are polymerized on plastic backing, have a size of 25 x 12.5 cm X 0.45 mm thick and are available with either 25 X 15 µL slots (25S) X or 52 slots X 6 µL (52 S) slots. Various gel concentrations are available: 10% *T*; 12.5% *T*; 15% *T* . The gels are backed using a non-fluorescent film specifically designed for fluorescent pre-labelling of proteins (DIGE) and/or fluorescent staining (LavaPurple). This backing is also suitable for traditional staining methods such as Coomassie or Silver. For long shelf-life and optimal separation a Tris-Glycine gel chemistry is used which maintains the pH of the gel is below 7.

Sample pre-treatment: Double the sample volume by adding an equal volume of sample buffer (2X) then dilute the sample to acheive the an appropriate gel loading concentration (this dends on the sensitivity of staining method used e.g. Coomassie Blue, Silver Staining or LavaPurple) using 1 X sample diluter. Then reduce and alkylate your sample.

1. Switch the thermostatic circulator on, set to 15 °C. Switch the FlatTop Tower on and set the valve to "*By-pass*" to avoid water condensation on the gel surface.
2. Lay two electrode wicks into the compartments of the PaperPool. Apply 20 mL of the respective electrode buffer to each wick and allow to soak for at least 10 minutes (Fig 1).
3. Apply at least 3 mL cooling contact fluid onto the cooling plate.
4. Remove the gel from its packaging. Remove the cover-film. Grip the gel (surface-up) at the two lateral edges at the protruding film, bend it like an "*U*" and slide the film-backing left and right on the cooling plate to distribute the cool contact fluid evenly (Fig. 2). Remove excess cooling fluid along film edges with lint-free tissue paper.
5. Place the cathode strip onto the cathodal edge of the gel. The edge of the strip should overlap the gel not more than by 2 mm. Place the anode strip on the anodal edge and remove any air by gentle rolling.

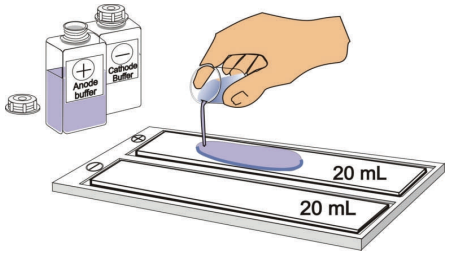

Fig. 1

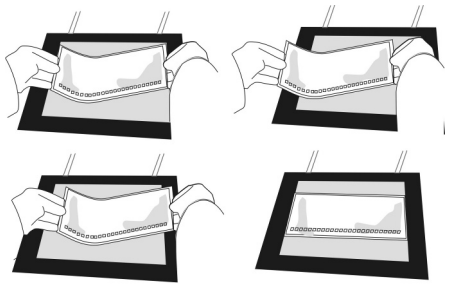

Fig. 2

6. Pipette 15 µL (25 S) or 6µL (52 S) of sample into the sample wells.
7. Clean platinum electrode wires before (and after) each electrophoresis run with moist tissue paper.
8. Close the lid while lowering the electrodes on the wicks, plug the cables in, turn the valve to cooling (15 °C)
9. Turn on your power supply and start the run according to table 1 or 2 depending on your sample type.

Table 1: Running conditions (15°C): Quick run for normal samples (total 2h)

Steps	Voltage	Current				Power				Time
		1 gel	***2 gels***	***3 gels***	***4 gels***	***1 gel***	***2 gels***	***3 gels***	***4 gels***	
S1	600 V ††	42 mA †	84 mA †	126 mA †	168 mA †	30 W ††	60 W ††	90 W ††	120 W ††	1 h
S2	1000 V ††	50 mA ††	100 mA ††	150 mA ††	200 mA ††	60 W †	120 W †	180 W †	240 †	1 h

Table 2 Running conditions (15°C): Slow run for difficult samples (total about 2 h 30 min)

S1	250 V ††	30mA †	60 mA †	90 mA †	120 mA †	10 W ††	20 W ††	30 W ††	40 W ††	45 min
S2	700 V ††	42 mA ††	84 mA ††	126 mA ††	168 mA ††	30 W †	60 W †	90 W †	120 W †	45 min
S3	1000 V ††	50 mA ††	100 mA ††	150 mA ††	200 mA ††	60 W †	120 W †	180 W †	240 W †	1 h

For programming BioRad Power Supplies only:

† - Set as "*constant*"

†† - Set as "*limit*"

Legal Information

Gelcompany Pty Limited,
Unit 3 43 – 51 College Street,
. Gladesville, NSW 2111, Australia.

Ordering Information

		Quantity	*gelcompany code no.*
Equipment:			
HPE-FlatTop Tower		1	PPT-001-TI
HPE-Thermostatic Circulator		1	PPC-100-TI
HPE-Power supply		1	PPP-001-TI
Accessories:			
HPE-FlatTop Tower Accessory Kit*			
*Comprising, 4 PaperPools, Scalpel handle, pack of scalpel blades, 2 forceps pointed, 2 forceps blunt, 4 Falcon tubes, scissors and roller		1 kit	PGT-001-TK
PaperPool (Tray for soaking the electrode strips)		4	1003-03
IPG Strip Equilibrator (Tray for IPG strip rehydration and equilibration)		1	1003-04
Steel Tray Standard (with grid, for hot Coomassie staining)		1	1003-25
Steel Tray Large (for large gels, with grid, for hot Coomassie staining)		1	1003-26
Steel Tray Multi 6 (for up to 6 large gels, with 6 grids)		1	1003-27
ScanFrame (for standard and large size gels)		1	PAS-001-TI
Cooling fluid			
HPE-FlatTop Cooling Fluid	50 mL	1	PCC-050-TI
	150 mL	1	PCC-150-TI

Gels and buffer kits (examples only - for other gel types please enquire)

1D SDS Electrophoresis - Standard size (25 x 12.5 cm) gels

Ready to use kits for 1D SDS-electrophoresis including cooling fluid, buffers and electrode wicks

10%*T*, 25 slots for 15µL (4 gels)	1 pack	PCS-125-TK
10%*T*, 52 slots for 6µL (4 gels)	1 pack	PCS-252-TK
15%*T*, 25 slots for 15µL (4 gels)	1 pack	PCS-325-TK
15% *T*, 52 slots for 6µL (4 gels)	1 pack	PCS-452-TK
12.5% *T*, 25 slots for 15µL on non-fluorescent film-backing* (4 gels)	1 pack	PCS-425-TK

*for DIGE-samples and other fluorescent staining

2D Electrophoresis Standard Size (25 x 12.5 cm) gels

HPE-2DGel flatbed NF Kits "Double gels":

Ready to use kits for running 2 X 11cm IPG-strips (*Double Gels*) including cooling fluid, buffers and electrode wicks.

2DGel flatbed NF 12.5%, 4 gels	1 pack	PCF-902-TK
2DGel flatbed NF 12.5%, 20 gels	1 pack	PCF-202-TK
2DGel flatbed NF 10-15% (gradient gels) 4 gels	1 pack	PCF-903-TK
2DGel flatbed NF 10-15% (gradient gels) 20 gels	1 pack	PCF-203-TK

HPE-2DGel flatbed NF Kits "Triple gels":

Kits for running 3 X 7 cm IPG-strips (*Triple Gels*) including cooling fluid, buffers, wicks and electrodes.

2DGel flatbed NF 12.5%, 4 gels	1 pack	PCT-301-TK
2DGel flatbed NF 12.5%, 20 gels	1 pack	PCT-302-TK
2DGel flatbed NF 10-15% (gradient gels) 4 gels	1 pack	PCT-321-TK
2DGel flatbed NF 10-15% (gradient gels) 20 gels	1 pack	PCT-322-TK

HPE-*Large* Format 2D Gels 25 X 19.5cm

Kits for running 1 X 24cm IPG-strips (Large Gels) including cooling fluid, buffers and electrode wicks.

2DGel flatbed NF 12.5%, 4 gels	1 pack	PCF-906-TK
2DGel flatbed NF 12.5%, 20 gels	1 pack	PCF-206-TK
2DGel flatbed NF 10-15% (gradient gels) 4 gels	1 pack	PCF-907-TK
2DGel flatbed NF 10-15% (gradient gels) 20 gels	1 pack	PCF-207-TK

Additives:

IPG-Ampholyte Mix, 1mL (40% w/v) for rehydration of IPG-strips.	1 pack	1004-15

Additionally Required (use only highest quality):

Urea, Dithiothreitol (DTT), Iodoacetamide (IAA), SDS marker proteins.

Fluorescent Stain Kit

Kit includes all fixatives and buffers required for fluorescently staining gels:

LavaPurple Protein Kit 100 mL	1 pack	LP-011100
LavaPurple Protein Kit 25 mL	1 pack	LP-011025

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