

Improved **Ettan** DALTsix Upper Buffer Chamber

Important information



Introduction

The improved Upper Buffer Chamber, in combination with updated protocols, is aimed at optimizing run results under the widest variety of running conditions.

Through ongoing research and development, the recommendations given below are designed to improve 2D results and extend the life time of the Upper Buffer Chamber (UBC) of the Ettan $^{\text{TM}}$ DALT six Electrophoresis Unit. Wherever possible please implement the following recommendations.

Preparing an electrophoresis run

Step Action

- When preparing to run a gel in the Ettan DALTsix
 Electrophoresis Unit, insert the anode assembly in the tank
 and then fill the lower buffer chamber tank with 4.5 liters of 1x
 electrophoresis buffer.
- 2 Turn on the circulation pump.
- 3 Connect an external MultiTemp™ III thermostatic circulator and set the temperature to 25°C for a day run and to 30°C for an overnight run.
- 4 Carefully insert the electrophoresis gels.
- Wet the UBC sealings with buffer solution or 0.1% SDS (immerse the sealings in solution or spray the sealings of the UBC using a plant sprayer) and carefully slide the UBC over the gel cassettes.

Do not move the UBC repeatedly up and down as this will reduce the sealing effect.

6 Fill the UBC with 1.2 liters of 2x buffer and use a funnel to adjust the buffer level in the lower buffer chamber to the same height as in the UBC, by adding 1x buffer.

Adjusting the buffer level

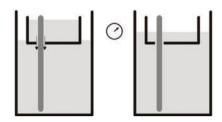


Fig 1. No Hydrostatic balance

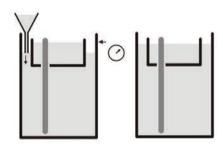


Fig 2. Hydrostatic balance

By adjusting the buffer level in the Lower buffer chamber to be the same as the level in the UBC, you achieve a status called 'hydrostatic balance', where there will be no overall change in the levels of either buffer chamber during electrophoresis. This will also help to improve runs and minimize any possible 'leakage' from the UBC.

Limitations and run conditions

The maximum rated electrical input for the Ettan DALTsix Electrophoresis Unit is 600 V 40 mA/gel and 13 W/gel. The maximum recommended temperature (on the MultiTemp III) is 30°C.

Run conditions (Day run):

Applicable to 1 mm thick 12% PAA gel and Laemmli Buffer system. Set the MultiTemp temperature to 25° C.

Step	mA/gel	Voltage (V)	W/gel	Time
				(hours:mins)
1	10	80	1	1:0
2	40	500	13	4:30-6:00 ¹

Continue the electrophoresis until the bromophenol blue reaches the end of the gel.

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Run conditions (Overnight):

Set the MultiTemp temperature to 30°C.

Step	mA/gel	Voltage (V)	W/gel	Time (hours:mins)
1	10	80	1	1:0
2	12	150	2	15:00-17:00 ¹

¹ Continue the electrophoresis until the bromophenol blue reaches the end of the gel.

Following the above running parameters will generate a temperature of around 30° C in the running buffer for day and night runs.

Running the electrophoresis at reduced power settings (as with few qels) will reduce the temperature differences accordingly.

Removing the gels

Step Action

- Remove the lid and carefully lift the UBC upwards making sure the cassettes remain in the anode assembly by applying downward pressure on them.
- 2 Lift the gel cassette holder upwards and remove the gels.

Care and Maintenance

Step Action

- On completion of the electrophoresis run, immediately remove the UBC from the Ettan DALTsix Electrophoresis Unit.
- Wash the UBC in distilled water of room temperature to wash off the buffer solution. Rinse thoroughly and allow the UBC to dry at room temperature.
- 3 Do not leave the UBC in buffer solution when the unit is not in use. Store the UBC empty, not even on the cassette holder, since the sealings in the middle may be deformed.

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