

PRODUCT INFORMATION

Excellent Gel Kit NF 12.5 % for 1D SDS-PAGE

Cat. No. 43423

Kit components:

4 pcs. 1D SDS TA Gel NF 12.5 % 25S

Buffer Kit:

250 ml SDS Anode Buffer (blue)

250 ml SDS Cathode Buffer (white)

8 pcs. Electrode wicks

Storage: +2 °C to +8 °C

Sample preparation:

1. Sample stock buffer:

- Dissolve 3.0 g Tris in 40 ml distilled water
- Adjust pH to 7.5 with approx. 1.4 ml acetic acid
- Make up to 50.0 ml with distilled water
- **Storage: 3 months at +2 °C to +8 °C**

2a. Sample buffer:

- 5.0 ml Sample stock buffer
+ 0.5 g SDS
+ 5 mg Bromophenol Blue
- Make up to 50 ml with distilled water and mix thoroughly
- **Storage: 1 month at +2 °C to +8 °C**

2b. Sample buffer (reducing):

- 5.0 ml Sample stock buffer
+ 0.5 g SDS
+ 77 mg DTT
+ 5 mg Bromophenol Blue
- Make up to 50 ml with distilled water and mix thoroughly
- **Use fresh solution daily**

Non-reducing SDS treatment

Dissolve the sample in sample buffer (2a) and heat at 95 °C for at least 3 min.

Reducing SDS treatment

Dissolve the sample in sample buffer (2b) and heat at 95 °C for at least 3 min.

Electrophoresis: Always wear powder free disposable gloves.

- Switch the thermostatic circulator on, set to 15 °C.
- Lay two electrode wicks into the compartments of the PaperPool. Apply 42 ml of the respective electrode buffer to each wick and allow soaking for at least 10 min (Fig 1).

- Apply 3 ml cooling contact fluid onto the cooling plate.
- 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.

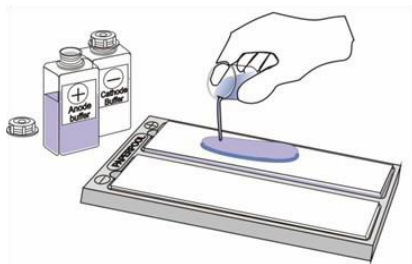


Fig. 1

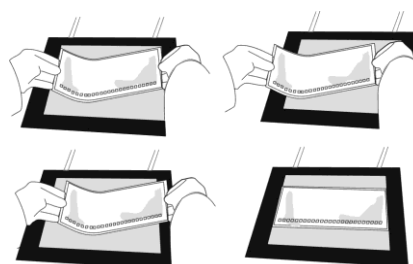


Fig. 2

- Remove excess electrode buffer from the wicks by tilting the electrode wicks along one long edge and dab it on the PaperPool bottom (fig. 3).

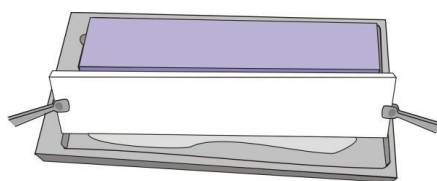


Fig. 3

- Place the electrode wicks onto the gel edges overlapping them by at least 2 mm. Hold wicks horizontally! Never sloped, because this would cause unequal buffer concentration along the wick. Smooth out air bubbles with bent tip forceps.
- Pipette **15 µl of sample** into the sample wells.
- Clean platinum electrode before (and after) each electrophoresis run with wetted tissue paper. Close the lid while lowering the electrodes on the wicks and plug the cables in.
- Turn on your power supply and start the run according to the used buffer system or for example as shown below (running conditions at 15 °C).

	Limit V	Limit mA	Set W	Time
1 Gel	600 V	42 mA	30 W	approx. 1 h 30 min