

Product Guide

VELUM Precast 1D IEF Gels

Product no. PR232-A - E, PR233-A - E, PR234-A - E

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1 Kit content

- 4 VELUM Precast 1D IEF Gels (5% AA/BisAA; size: 250 x 110 x 0.67 mm)
- VELUM Precast Gel Cooling Solution

2 Additional material required (not included)

- materials required for sample preparation
- ORCA Gel Electrophoresis System or horizontal electrophoresis system

3 Storage and stability

See package label

4 Gel types

Product no.	pH gradient	Slots	Slot volume	suitable for (fluorescence)detection
PR 232-A	3-10	25	20 µl	red and infrared, Coomassie
PR 232-B	4-6	25	20 µl	red and infrared, Coomassie
PR 232-C	2-7	25	20 µl	red and infrared, Coomassie
PR 232-D	4-5	25	20 µl	red and infrared, Coomassie
PR 232-E	6-11	25	10 µl	red and infrared, Coomassie
PR 233-A	3-10	40	10 µl	red and infrared, Coomassie
PR 233-B	4-6	40	10 µl	red and infrared, Coomassie
PR 233-C	2-7	40	10 µl	red and infrared, Coomassie
PR 233-D	4-5	40	10 µl	red and infrared, Coomassie
PR 233-E	6-11	40	10 µl	red and infrared, Coomassie
PR 234-A	3-10	80	8.5 µl	red and infrared, Coomassie
PR 234-B	4-6	80	8.5 µl	red and infrared, Coomassie
PR 234-C	2-7	80	8.5 µl	red and infrared, Coomassie
PR 234-D	4-5	80	8.5 µl	red and infrared, Coomassie
PR 234-E	6-11	80	8.5 µl	red and infrared, Coomassie

5 General information

VELUM Precast Gels are designed for high resolution horizontal protein separation of complex protein samples. Due to a film-backing support VELUM Precast Gels do not require glass plates and run with very small amounts of buffer.

6 Instructions for use

6.1 Preparation of the horizontal electrophoresis system

- Clean the cooling plate and the electrodes with ddH₂O using a lintfree tissue.
- Apply up to 3 ml VELUM Precast Gel Cooling Solution onto the cooling plate along the center line.

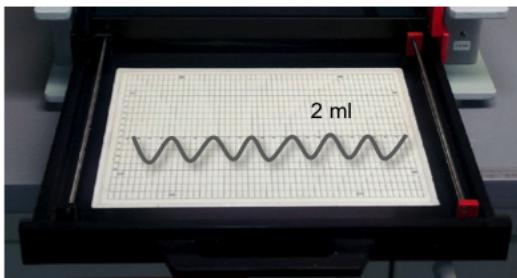


Fig. 1 Application of Cooling Solution

6.2 Placing the Gel

- Remove the upper cover film (larger than the gel) from the VELUM Precast Gel
- Place the gel with the gel film support downwards and the wells towards you onto the VELUM Precast Gel Cooling Solution.

Caution: Avoid air bubbles between gel and cooling plate.

- Remove remaining VELUM Precast Gel Cooling Solution using lint-free tissues.

6.3 Pre-Focusing (optional)

- For some sample types it is beneficial to start with pre-focusing of the pH gradient before sample application.
- use the following setting for pre-focussing: 1000 V, 25 mA, 10 W, 20 min

6.4 Sample application

- Apply appropriate volume of sample into the gel slots. Maximum load capacity: see 4. Gel Types.
- for a small number of samples halve the gel.

Note: If you are using one half of a gel only, adapt electrophoretic settings by reducing the current (mA) and power (W) by 50%. Use the voltage (V) and time settings as indicated in 6.6.

6.5 Placing the electrodes

ORCA Gel Electrophoresis Unit:

Place each of the cleaned platinum electrodes directly onto the gel surface at the outer edges of the gel. Press the platinum electrodes onto the gel to ensure optimal contact.

6.6 Isoelectric focussing

- Switch on the Cooling Unit, adjust to 7 °C.

- Start the IEF.
- use the following parameters for one gel:

VELUM Precast 1D IEF Gels (0.65 mm): ca. 2,5 h			
Parameter	Step 1	Step 2	Step 3
Voltage	500 V	1650 V	1800 V
Current	8 mA	22 mA	22 mA
Power	10 W	15 W	20 W
Time	30 min	90 min	30 min
Actual start voltage (appr.)	60 V	750 V	1800 V
Temperature	7°C		

For a half gel: halve mA and W, do not change V !

- After electrophoresis remove the gel from the electrophoresis unit and rinse the gel using ddH₂O to remove Cooling Solution. Proceed to Post-electrophoretic application (Staining, Imaging, Blotting etc.).
- Clean the Cooling plate of the horizontal electrophoresis system and the electrodes with ddH₂O. For cleaning use lintfree tissues.

7 Fixation and Storage of the gel

Fixate proteins in the gel for 30 min using 20% (w/v) TCA. Store gels in 20% (v/v) Ethanol and 3% (v/v) glycerol.

8 Staining / Imaging / Blotting

VELUM Precast 1D IEF Gels are suitable for red and infra-red fluorescence detection, Coomassie staining or blotting onto PVDF or Nitrocellulose transfer membranes.

Avoid concentrations of organic solvents above 40% (v/v).

For Blotting of the VELUM Precast Gels use the BEO or VELUM Dry Blotter (PR 87 und PR88).