Userguide

epMotion 5070/5075 | AU006

Loading the Invitrogen E-Gels with the epMotion 5070/5075

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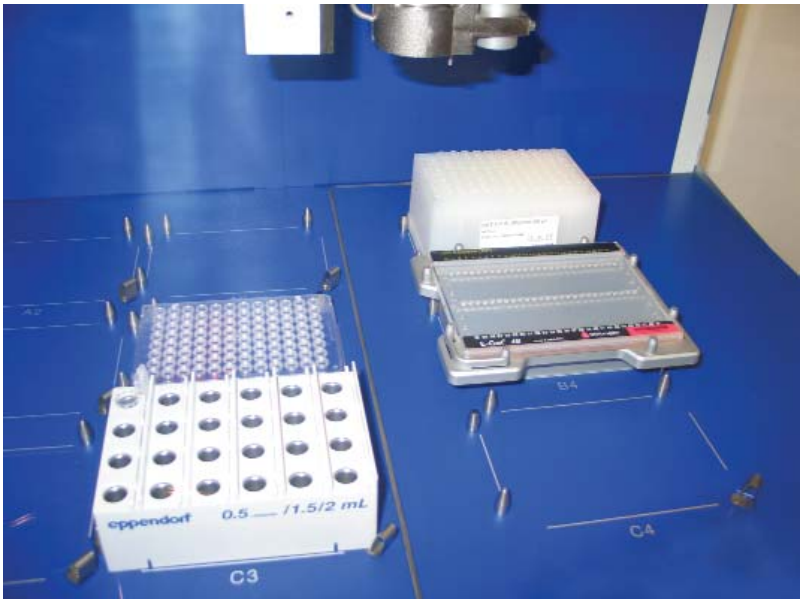

Fig. 1: The worktable of epMotion 5075 LH with the labware for loading of the E-Gel 48.

Introduction

Eppendorf as a recognised provider of liquid handling equipment and automatic systems has developed an array of state-of-the-art products enabling a full automation and streamlining of many routine laboratory applications. One of the basic protocols performed in the daily laboratory practice is the nucleic acid separation and evaluation by the agarose gel electrophoresis. We report here two recently developed protocols for the automated agarose gel loading employing the Eppendorf workstation epMotion 5075® and the Invitrogen E-Gel® system in 48 and 96 formats.

Methods and Results

General Considerations

The E-Gels work together with the specifically developed holder (E-Gel Holder), which base is of the standard SBS plate format. For the optimal loading procedure we recommend using the epMotion 5075 LH and positions A4, B4 and C4. The other epMotion models: 5070 and 5075 VAC and 5075 MC provide less free space and can be used with precautions to sterical hindrance between E-Gel/Holder and other labware (E-Gels are bigger than SBS format and in certain labware combinations may require two positions on the epMotion worktable).

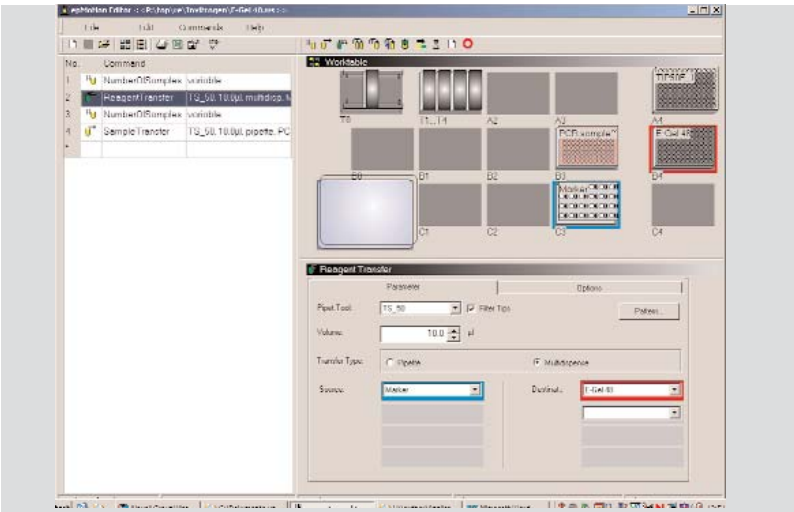

Fig. 2: epMotion Editor screenshot of the epMotion 5075 LH worktable with the labware for loading the E-Gel 48.

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Loading the gels
Invitrogen Holder; Cat. no. G7300-01

Loading the E-Gels in 48 format
Invitrogen E-Gel 48, 2% Agarose (GP); Cat. no. G8008-02
Invitrogen E-Gel 48, 4% Agarose (GP); Cat. no. G8008-04

Labware file: epMotion /Plates/PCR96/INV_egel_48
epMotion-Method: E-Gel 48.ws

Below we provide an exemplary protocol: 48 PCR samples are transferred from the PCR plate (on a Thermoadapter) to the E-Gel 48 using a single channel tool TS-50 (Sample Transfer command). The additional 1,5 ml tube with the molecular weight marker was provided on a separate 24 Tube Thermorack. Please note that the epMotion control panel software allows only the display of 24 out of 26 wells of the E-Gel 48. The omitted wells are nonetheless included in the procedure. The markers must therefore be programmed within the visible area of the pattern.

The layout of the E-Gel 48 allows the usage of the single-channel tool only. The labware file of the gel is configured in such a way that no further adjustment is necessary for loading the gel. If the distance between the pipette tip and the gel should nonetheless be reduced, as with other plates, the bottom tolerance can be edited (3). Please note: the liquid level detection option is not possible and optical sensor should be switched off for the E-Gel work position.

Table 1: Detailed listing of the labware required for loading the E-Gels 48.

Worktable Position	Labware	Comment
T0	-	
T1 - 4	Dispensing tool TS 50	
A2	-	
A3	-	
A4	ep T.I.P.S. Motion 50 µl	48 tips for 48 samples
B0	-	
B1	-	
B2	-	
B3	Thermoadapter 96 Eppendorf twin.tec PCR plate	Thermoadapter for PCR plates Samples found prepared in the PCR plate
B4	Invitrogen holder and Invitrogen E-Gel 48	Invitrogen E-Gel 48, 2% Agarose
C1	-	
C2	-	
C3	Thermorack 24	1.5 ml Safe-Lock Tubes filled with marker
C4	-	

Table 2: The E-Gel 48 loading protocol

1	Number of Samples	Variable, max. 4
2	Reagent Transfer	Transfer of the marker into the appropriate gel pockets
3	Number of Samples	Variable, max. 48
4	Sample Transfer	Transfer of the samples into the provided gel pockets

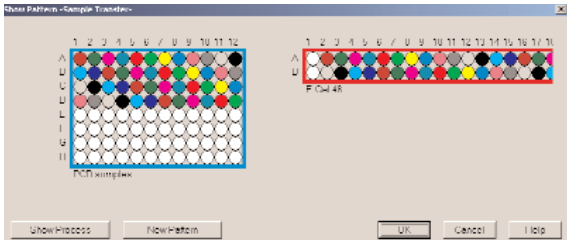

Fig. 3: The epMotion Editor screenshot version 9.22: example of loading pattern for E-Gel 48: 18 x 2 gel positions are visible. Please note: the epMotion control panel software makes 24 x 2 positions visible (not shown here).

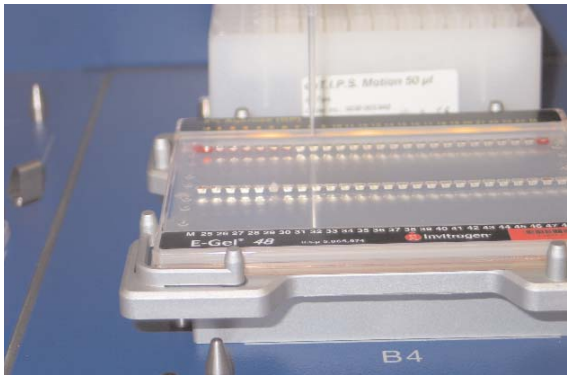

Fig. 4: Loading of the E-Gel 48 with the single-channel dispensing tool TS-50.

E-Gel in 96 format
Invitrogen E-Gel 96, 1% Agarose (GP); Cat. no. G7008-01
Invitrogen E-Gel 96, 2% Agarose (GP); Cat. no. G7008-02

Labware file:
epMotion/Plate/PCR96/
InV_e_gel_96_8_K
epMotion-Method: E-Gel 96(8 channel).ws

Exemplary protocol for the E-Gel 96 is provided below: 96 PCR samples are transferred from the PCR plate (on a Thermoadapter) to the E-Gel using the eight-channel tool TM-50 (Sample Transfer command).

The molecular weight marker was provided in a 1,5 ml Safe-Lock tube in a separate Thermorack and transferred with the Reagent Transfer command using the single–channel tool TS-50.
The dispensing distance of the pipette tips above the gel can be varied by editing the bottom tolerance (3).

After the loading procedure the E-Gels are removed from the E-Gel holder and used for electrophoresis in the provided E-Gel Mother Base. For further information please refer to the Invitrogen E-Gel manual.

Summary

The automated E-Gel loading protocols reported here provide the last step in the numerous nucleic acid applications. Combined with the automated PCR reaction setup, PCR cleanup, as well as genomic and plasmid DNA purification they offer a comprehensive solution for a complete lab automation.

Table 3: Detailed listing of the labware required for the E-Gel 96 loading

Position	Labware	Comment
T0	-	
T1 - 4	Dispensing tool TM 50	
A2	-	
A3	-	
A4	ep T.I.P.S. Motion 50 µl	96 tips for 48 samples
B0	-	
B1	-	
B2	-	
B3	Thermoadapter 96 Eppendorf twin.tec PCR plate	Thermoadapter for PCR plates Prepared samples in the PCR plates
B4	Invitrogen holder and Invitrogen E-Gel 96	Invitrogen E-Gel 96, 1% Agarose
C1	-	
C2	-	
C3	Thermorack 24	1.5 ml Safe-Lock Tubes filled with marker
C4	-	

Table 4: E-Gel 96 loading protocol

1	Number of Samples	Variable, max. 8
2	Reagent Transfer	Transfer of the marker into the appropriate gel pockets
3	Number of Samples	Variable, max. 96
4	Sample Transfer	Transfer of the samples into the provided gel pockets

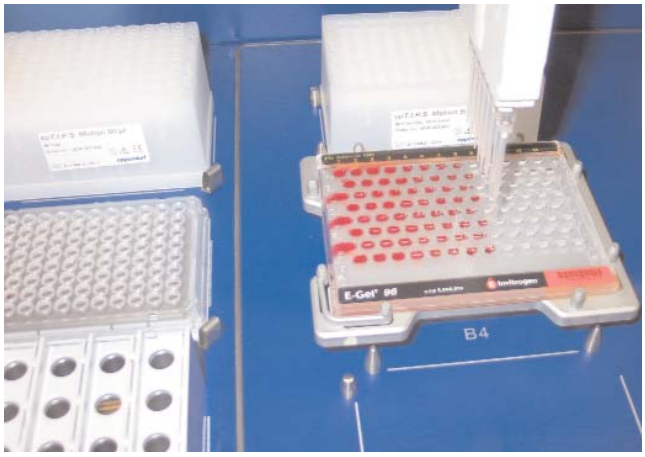

Fig. 7: Filling of the gel with the 8-channel dispensing tool

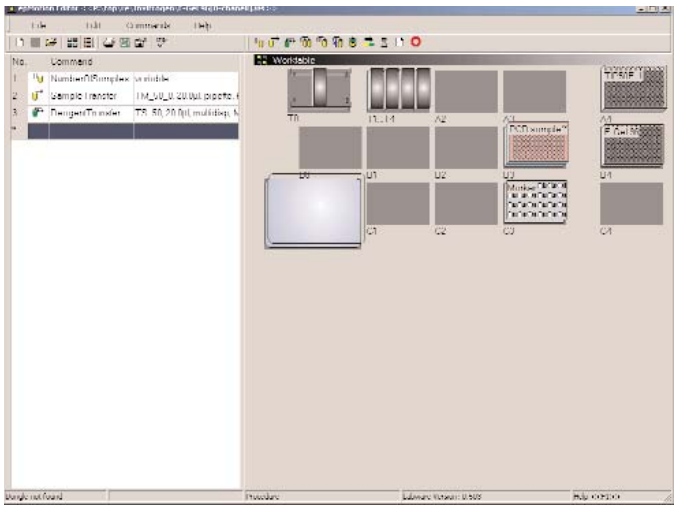

Fig. 5: epMotion Editor screenshot of the epMotion 5075 LH worktable with the labware for loading the Invitrogen E-Gel 96.

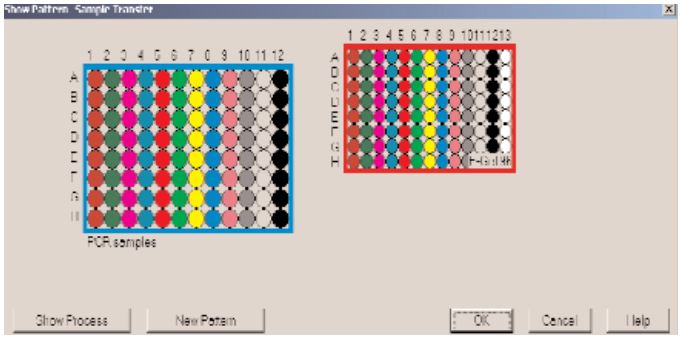

Fig. 6: The epMotion Editor screenshot: example of loading pattern for E-Gel 96. The 13th row remains free for the markers (pipetted by Reagent Transfer command with the single-channel tool TS-50).

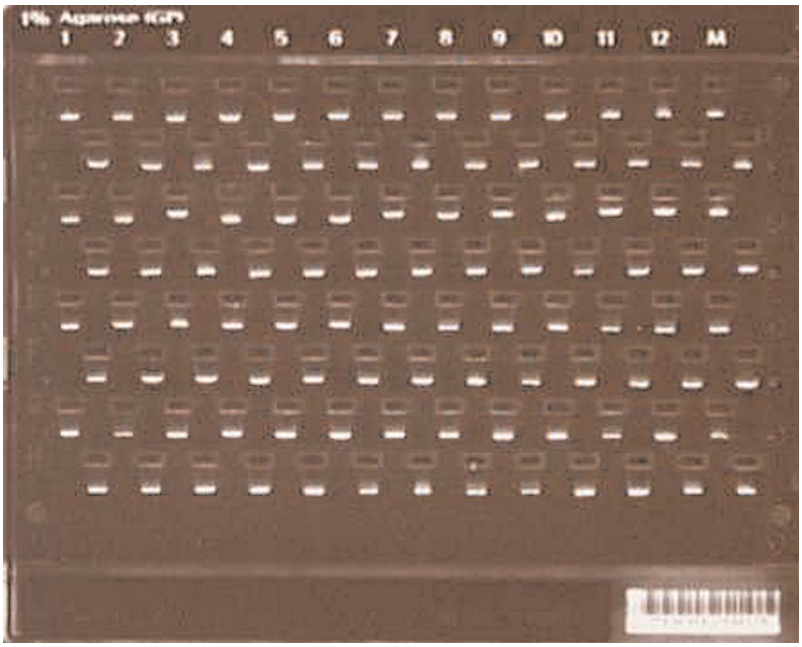

Fig. 8: 96 x 20 µl plasmid DNA samples were loaded on a 1% Invitrogen E-Gel 96 with the epMotion 5075 LH. Electrophoresis was performed on the E-Gel Mother Base.

References

1. Instruments Manual for the epMotion 5070 or epMotion 5075 Workstations
2. Instruction Manuals for Invitrogen E-Gel 48 or Invitrogen E-Gel 96
3. Userguide epMotion 5070/5075 AU005

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