

Instructions for use

Horizontal Electrophoresis Units

2788.1 ROTIPHORESE® Unit PROfessional I

2799.1 ROTIPHORESE® Unit PROfessional II

2850.1 ROTIPHORESE® Unit PROfessional III

9938.1 ROTIPHORESE® Unit PROfessional III Stretch 20

9939.1 ROTIPHORESE® Unit PROfessional III Stretch 25

2941.1 ROTIPHORESE® Unit PROfessional IV

3000.1 ROTIPHORESE® Unit PROfessional V

<u>Warning</u>: Like all apparatus run by electricity these units are capable of delivering potentially lethal voltage when connected to a power supply. They should be operated only by qualified technically trained personnel.

The horizontal electrophoresis units from ROTH are designed for long term laboratory use and to obtain reproducible results. Please spend a few moments reading the instruction manual thoroughly.

These units comply with the statutory CE safety rules:

73/23/EEC: Low voltage directive: IEC 1010-1:1990 plus amendment 1:1992

EN 61010-1:1993/BS EN 61010-1:1993

Please verify, that you received the unit completely and without any damage. Any faults or losses have to be reported to ROTH immediately. ROTH can not accept responsibility for goods, that were sent back without informing them.

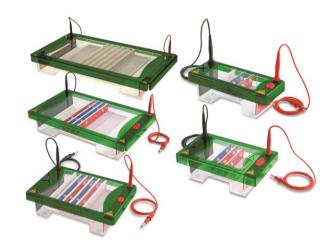
Please take a look at the packing list and check whether all components and accessories are present

Please retain all packaging material until the warranty period has expired.

SPECIFICATION

Technical features:

- Injection moulded acrylic construction
- Non-sliding rubber base
- Very easily replaceable electrodes
- Doubly insulated cables
- o Gold plated electrical connectors, corrosion-free
- Recessed power connectors
- 0.2 mm diameter platinum electrodes, 99.99 % pure
- Removable UV transparent gel casting trays
- Large variation of gel casting trays
- Combs colour coded: 0.75 mm black,
 1.0 mm white, 1.5 mm red, 2.0 mm blue
- o Combs adjustable in height



Environmental Conditions

- This apparatus is intended for indoor use only.
- The unit can be operated safely at an altitude of 2000 m.
- The normal operating temperature range is between 4 °C and 65 °C.
- Maximum relative humidity 80 % for temperatures up to 31 °C decreasing linearly to 50 % relative humidity at 40 °C.

All Roth products are supplied having passed rigorous quality control procedures. For additional questions please give us a call: ++49-721-5606-0

PACKING LIST

Art. No.	Unit	Gel Casting Tray I x w (cm)	Gel Casting Gates	Combs
2788.1	PROfessional I	1 (7 x 7)	2	2 (1 mm, 8 teeth)
2799.1	PROfessional II	1 (10 x 10)	2	2 (1 mm, 16 teeth)
2850.1	PROfessional III	1 (15 x 15)	2	2 (1 mm, 20 teeth)
9938.1	PROfessional III Stretch 20	1 (15 x 20)	2	4 (1 mm, 28 teeth)
9939.1	PROfessional III Stretch 25	1 (15 x 25)	2	4 (1 mm, 28 teeth)
2941.1	PROfessional IV	1 (20 x 20)	2	2 (1 mm, 20 teeth)
3000.1	PROfessional V	1 (26 x 32)	0	6 (1 mm, 28 teeth)

Tab.1 Operational:

Art. No.	normal operating voltage (V)	Normal running time (min)	normal operating current (mA)	approx. gel volume (ml)	approx. buffer volume (ml)
2788.1	35 – 80	45 – 60	50	25	225
2799.1	50 – 100	45 – 60	75	50	300
2850.1	90 – 150	60 – 90	100	112,5	500
9938.1	100 – 150	60 – 90	100	150	1000
9939.1	100 – 150	60 – 90	100	187,5	1000
2941.1	100 – 150	60 – 90	100	200	1500
3000.1	100 – 150	90 – 120	100	416	2000

USING THE HORIZONTAL GEL ELECTROPHORESIS UNITS

A. Safety Precautions

- Please read the entire instruction manual thoroughly before using the apparatus.
- Always isolate electrophoresis units from their power supply before removing the safety cover.
 Isolate the power supply from the mains <u>first</u> then disconnect the leads.
- Do not exceed the maximum operating voltage or current (see table 1).
- Please wear always protective gloves while working.
- Do not fill the unit with running buffer above the maximum fill lines.
- Do not move the unit when it is running.

CAUTION: During electrophoresis very low quantities of various gases are produced at the electrodes. The type of gas produced depends on the composition of the buffer employed. To disperse these gases, make sure that the apparatus is run in a well ventilated area.

B. General Care and Maintenance

Clean the apparatus with hand warm water and a mild detergent only. Often, a thorough rinse with distilled water is all that is required. Dry components with clean tissues prior to use, e.g. ROTH tissues (ref. 0087.2)

Important: Acrylic plastic is not resistant to aromatic or halogenated hydrocarbons, ketones, esters, alcohols (over 25 %) and acids (over 25 %), they will cause "crazing" of the plastic and should not be used for cleaning. Do not use abrasive creams or scourers. The units should never come into contact with the following cleaning agents, these will cause irreversible and accumulative damage: Acetone, Phenol, Chloroform, Carbon tetrachloride, Methanol, Ethanol, Isopropyl alcohol, Alkalis.

Before use, and then on a monthly basis, check the unit for any leaks at the bonded joints. Place the unit on a sheet of dry tissue and then fill with distilled water only to the maximum fill line. If any leakage is seen do not attempt to repair or use the apparatus, but notify Carl Roth GmbH & Co. KG immediately (+49/0721/5606-172).

The replacement platinum electrodes are partially shrouded for protection.

However, when cleaning the main tank do not use cleaning brushes in the electrode area.

Ensure that the connectors are clean and dry before usage or storage.

RNase Decontamination

Clean the units with a mild detergent as described above.

Wash with 3% hydrogen peroxide (H₂O₂) for 10 minutes.

Rinsed with 0.1% DEPC- (diethyl pyrocarbonate) treated distilled water,

Caution: DEPC is a suspected carcinogen. Always take the necessary precautions when using.

ROTI® Nucleic Acid free (Art. No. HP69) and RNAse AWAY^(TM) (Art. No. A998) may also be used. Please consult the instructions for use with acrylic gel tanks.

C. Preparations

Fitting Electrode Cables

- 1. Note the position of the lid on the unit. This shows the correct polarity and the correct orientation of the cables, black is negative and red positive.
- 2. Remove the lid from the unit. Note if the lid is not removed, fitting the cables may result in untightening of the gold plug and damage to the electrode.
- 3. Screw the cables into the tapped holes as fully as possible so that there is no gap between the lid and the leading edge of the cable fitting.
- 4. Refit the lid.

Fitting Loading Guides

These can be fitted to enhance visibility of the wells if desired. They can be fitted to the white vinyl platform sheet or to the unit itself.

- 1. Seat the tray in the unit and note the position of the comb grooves. The samples run black to red but the trays can be used frontward or backwards so ensure that the comb grooves closest to the black electrode are marked.
- 2. Remove the tray.
- 3. Peel the back off of the loading guide and carefully apply the loading guide directly to the gel platform.

D. Gel Pouring

1. The following amounts of agarose are needed to prepare a gel of 5 mm height:

PROfessional		II	Ш	III-20	III-25	IV	V
Gel size	7 x 7	10 x 10	15 x 15	15 x 20	15 x 25	20 x 20	26 x 32
Agarose (ml)	25	50	112,5	150	187,5	200	416

2. Fit the casting dams over each end of the tray and place onto a level surface. The dams should be fitted so that there is no gap between the sides of the tray and the groove in the dams. This will ensure that there is no possibility of gel leakage.

The PROfessional V does not contain casting dams. Please use traditional tape for **sealing of the gel casting tray**. We recommend use of the temperature resistant SEKUROKA®® tape (Art. No. L035.1). Alternatively, you can use the external gel casting stand PROfessional Flexicaster (Art. No. 3688.1). For further description see page 6 in this manual.



- 3. Place the gel casting tray on a flat surface or use the Roth levelling table (Art. No. N854.1).
- 4. Insert the appropriate comb into the grooves. Cool the agarose down to 50-60 °C in order to avoid leaking of the gel during pouring and any damage to the gel tray. Pour the agarose to the desired height (approx. 5 mm).
- 5. Do not move the casting tray until the gel has polymerised. We recommend further polymerisation of the agarose gel for approx. 10 min in the refrigerator.
- 6. Remove the casting dams carefully.
- 7. Place the gel casting tray and agarose gel into the electrophoresis chamber and submerse the gel in running buffer. Remove the comb carefully.

E. Running the Gel

Because of the numerous applications now being used in horizontal gel electrophoresis, no specific running conditions are given. As a guide to obtain the optimum resolution of DNA fragments, agarose gels should not be run greater than 5 V / cm. Specific protocols are available in the numerous laboratory handbooks and publications.

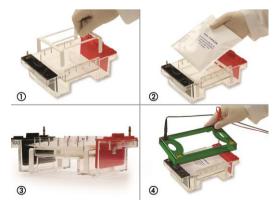
Add gel loading buffer to the DNA-samples and load the samples into the wells. Replace the lid correctly before connecting the leads to the power supply.

Set the voltage and current to suit the electrophoretic application.

Important: Do not exceed the recommended voltage or current as this may result in poor band resolution and may result in damage to the unit. More information provides **Table 1 "Operational"** in this manual.

Long runs or runs under higher voltage may require cooling. Precool the cooling pack at -20 °C for several hours and then place it on top of the loaded gel using the cooling frame. If required, cooling packs may be exchanged during the run.

NOTE: Make sure to take of current and disconnect the electrophoresis unit from Power Supply before opening the unit!



F. At the End of the Run

Turn the power supply settings to zero, turn off mains supply and disconnect the power leads.

Remove safety lid at the end of the run and take gel tray out of the chamber for staining. Since all gel casting trays are UV-permeable, staining can occur without removing the gel from the tray.

Rinse the apparatus with distilled water only after the run (see section: **B General Care and Maintenance**).

Ensure that the connectors are clean and dry before usage or storage.

G. Additional Items and Reagents

Agaroses: Standard NEEO ultra-quality Agarose-Tablets Broad Range (for all fragment lengths) GTQ (gene technique quality − for DNA elution) Agarose HR-PLUS (for fragm. of 100 − 3000 bp) High Resolution (for small fragment lengths) Low Melt (for gel elution and in-gel-applications) () Agarose LM/PCR (Gel elution of fragm. < 1500 bp) Agarose Super LM (particularly low melting temp.) Synergel™ (Agarose additive for even better band resolution)	3810 2267 HP67 T846 6352 HP30 K297 6351 HP31 HP54 0184
Gel Loading Buffers: ROTI®Load DNA 6x (with Glycerol / Ficoll) ROTI®Load DNA short run 6x (with Glycerol) ROTI®Load DNA 1x (with Glycerol) ROTI®Load DNA short run 1x (with Glycerol) ROTI®Load DNA small (with Glycerol) ROTI®Load DNA orange 1 (with Glycerol) ROTI®Load DNA orange 2 (with Glycerol) ROTI®Load DNA tricolor (with Glycerol) ROTI®Load DNAstain 1 SYBR® Green (for fragments > 500 bp) ROTI®Load DNAstain 2 SYBR® Green (for fragments < 500 bp) ROTI®Load DNAstain 3 SYBR® Green (for fragments < 500 bp)	X904 / X905 0095 0100 0099 HP03 HP04 HP05 HP06 1CN5 1CN6 1CN7
Gel Running Buffers: ROTIPHORESE®- 10 x TBE-Buffer ROTIPHORESE®- 10 x TAE-Buffer ROTIPHORESE®- 10 x TAE-Buffer light (for gel elution)	3061 T845 0122
DNA Markers: Please call ++49-0721-5606-0 for our brochure on DNA Markers	;
Staining reagents: Ethidium bromide solution 1 % Ethidium bromide solution 0.5 % in dropper bottle Ethidium bromide solution 0.025 % in dropper bottle Ethidium bromide dye ROTI®GelStain (green fluorescent dye, substitute for eth. bromide) ROTI®GelStain Red (red fluorescent dye, substitute for eth. bromide) SYBR® Green DNA dye (green fluorescent dye, substitute for eth. bromide) Methylene blue staining solution Methylene blue dye	2218 HP46 HP47 7870 3865 0984 1CN2 0648 A514
SEKUROKA® Decon Bags (for removal of 125 mg eth. bromide from solutions)	T856

3688.1 ROTIPHORESE® PROfessional Flexicaster

External Gel Casting Unit

For use with the electrophoresis units PROfessional I-V

Casting unit for pouring gels outside the gel chamber, suitable for all PROfessional gel trays up to a size of 26 x 32 cm (W x L).

When using gel trays up to a size of 10 x 10 cm, 2 gels can be poured simultaneously (PROfessional I and II).

The level bubble allows an optimal levelling of the gels. Additional sealing is not necessary.



N854.1 Levelling Table

Carl ROTH levelling tables enable casting of horizontal gels of equal thickness, featuring easy-to-turn rotating feet on the corners.

Tables also have an air bubble in the center for precise horizontal alignment.



H. Combs

2788.1 ROTIPHORESE® Unit PROfessional I

Sample volume for a 5 mm thick gel											
	1 + 1	2 + 2	4 + 2	8	10	12	16				
0.75 mm	152 µl	68 µl	36 µl	19 µl	14 µl	10 µl	7 µl				
1.0 mm	203 µl	90 µl	48 µl	25 µl	18 µl	14 µl	10 µl				
1.5 mm	304 µl	135 µl	72 µl	37 µl	27 µl	20 µl	15 µl				
2.0 mm	405 µl	180 µl	96 µl	50 µl	36 µl	27 µl	20 µl				

Wells	1 + 1	2 + 2	4 + 2	8	10	12	16
Thickness	Art. No.	Art. No.	Art. No.	Art. No	Art. No.	Art. No.	Art. No.
0.75 mm	2737.1**	2738.1**	2739.1**	2740.1	2741.1	2742.1*	2743.1
1.0 mm	2744.1**	2745.1**	2746.1**	2747.1	2748.1	2749.1*	2750.1
1.5 mm	2751.1**	2752.1**	2753.1**	2754.1	2755.1	2756.1*	2757.1
2.0 mm	2758.1**	2759.1**	2760.1**	2761.1	2762.1	2763.1*	2764.1

^{*}compatible with multi-channel pipettors

^{**}comb with especially broad wells + narrow marker well (for preparative gels)

2799.1 ROTIPHORESE® Unit PROfessional II

	Sample volume for a 5 mm thick gel											
	2 + 2	4 + 2	8	10	12	16	20					
0.75 mm	118 µl	57 µl	30 µl	20 µl	17 µl	12 µl	10 µl					
1.0 mm	158 µl	77 µl	41 µl	27 µl	23 µl	16 µl	14 µl					
1.5 mm	236 µl	115 µl	61 µl	41 µl	34 µl	24 µl	20 µl					
2.0 mm	315 µl	153 µl	81 µl	54 µl	45 µl	32 µl	27 µl					

Wells	2 + 2	4 + 2	8	10	12	16	20
Thickness	Art. No.						
0.75 mm	3150.1**	3151.1**	3157.1	3158.1*	3161.1	3177.1	3178.1*
1.0 mm	3181.1**	3188.1**	3202.1	3203.1*	3204.1	3206.1	3208.1*
1.5 mm	3212.1**	3213.1**	3219.1	3220.1*	3222.1	3227.1	3229.1*
2.0 mm	3230.1**	3233.1**	3234.1	3238.1*	3239.1	3244.1	3245.1*

^{*}compatible with multi-channel pipettors

2850.1 ROTIPHORESE® Unit PROfessional III 9938.1 ROTIPHORESE® Unit PROfessional III Stretch 20 9939.1 ROTIPHORESE® Unit PROfessional III Stretch 25

	Sample volume for a 5 mm thick gel										
	4 + 2 10 12 16 20 28 32										
0.75 mm	91 µl	34 µl	30 µl	20 µl	16 µl	8 µl	7 µl				
1.0 mm	122 µl	45 µl	41 µl	27 µl	21 µl	11 µl	10 µl				
1.5 mm	182 µl	68 µl	61 µl	41 µl	32 µl	17 µl	15 µl				
2.0 mm	243 µl	90 µl	81 µl	54 µl	43 µl	23 µl	20 µl				

Wells	4 + 2	10	12	16	20	28	32
Thickness	Art. No.						
0.75 mm	3263.1**	3266.1	3269.1	3270.1*	3276.1	3281.1*	3283.1
1.0 mm	3302.1**	3308.1	3309.1	3312.1*	3317.1	3332.1*	3334.1
1.5 mm	3339.1**	3347.1	3350.1	3365.1*	3368.1	3370.1*	3372.1
2.0 mm	3373.1**	3374.1	3378.1	3379.1*	3381.1	3382.1*	3384.1

^{*}compatible with multi-channel pipettors

2941.1 ROTIPHORESE® Unit PROfessional IV

	Sample volume for a 5 mm thick gel											
	4 + 2	4 + 2 16 20 25 30 36 40 50										
0.75 mm	115 µl	30 µl	20 µl	16 µl	13 µl	11 µl	8 µl	8 µl				
1.0 mm	153 µl	41 µl	27 µl	21 µl	17 µl	14 µl	11 µl	10 µl				
1.5 mm	230 µl	61 µl	41 µl	32 µl	26 µl	22 µl	17 µl	16 µl				
2.0 mm	306 µl	81 µl	54 µl	42 µl	34 µl	29 µl	23 µl	21 µl				

Wells	4 + 2	16	20	25	30	36	40	50
Thickness	Art. No.							
0.75 mm	3401.1**	3409.1	3410.1*	3413.1	3414.1	3416.1	3417.1*	3419.1
1.0 mm	3422.1**	3427.1	3428.1*	3430.1	3431.1	3432.1	3435.1*	3437.1
1.5 mm	3438.1**	3440.1	3441.1*	3442.1	3443.1	3444.1	3445.1*	3446.1
2.0 mm	3447.1**	3449.1	3450.1*	3452.1	3453.1	3454.1	3456.1*	3458.1

^{*}compatible with multi-channel pipettors

^{**}comb with especially broad wells + narrow marker well (for preparative gels)

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3000.1 ROTIPHORESE® Unit PROfessional V

Sample volume for a 5 mm thick gel				
	28	56		
0.75 mm	25 µl	10 µl		
1.0 mm	34 µl	14 µl		
1.5 mm	51 µl	20 µl		
2.0 mm	68 µl	27 µl		

Wells	28	56
Thickness	Art. No.	Art. No.
0.75 mm	3023.1*	3024.1*
1.0 mm	3027.1*	3031.1*
1.5 mm	3038.1*	3055.1*
2.0 mm	3056.1*	3058.1*

I. Accessories

System	Gel trays (without casting dams)		Gel casting
			dams (2)
	Gel size (B x L)	Art. No.	
PROfessional I	7 x 7 cm	2784.1	2765.1
PROfessional I	7 x 10 cm	2785.1	2765.1
PROfessional II	10 x 7 cm	3147.1	3146.1
PROfessional II	10 x 10 cm	3148.1	3146.1
PROfessional III	15 x 7 cm*	3250.1	3262.1
PROfessional III	15 x 10 cm*	3251.1	3262.1
PROfessional III	15 x 15 cm*	3252.1	3262.1
PROfessional III Stretch 20	15 x 20 cm	9940.1	3262.1
PROfessional III Stretch 25	15 x 25 cm	9941.1	3262.1
PROfessional IV	20 x 10 cm	3392.1	3397.1
PROfessional IV	20 x 20 cm	3393.1	3397.1
PROfessional IV	20 x 25 cm	3394.1	3397.1
PROfessional V	26 x 10 cm	3019.1	-
PROfessional V	26 x 20 cm	3020.1	-
PROfessional V	26 x 32 cm	3021.1	-

^{*}compatible with PROfessional III Stretch units

	Gel cooling	Positive electrode	Negative electrode
PROfessional I	-	2767.1	2769.1
PROfessional II	-	3141.1	3142.1
PROfessional III	-	3247.1	3248.1
PROfessional III Stretch 20	-	3247.1	3248.1
PROfessional III Stretch 25	-	3247.1	3248.1
PROfessional IV	3386.1	3390.1	3391.1
PROfessional V	3009.1	3001.1	3002.1

J. Trouble shooting

Gel leaks during casting

- Please check that the rubber seals on the gel casting tray and gel casting gates are tight.

Air bubbles in gel during casting

- Remove the bubbles **immediately** with a comb or spatula.
- Ensure that the agarose is boiled well before casting to degas it. In the event of a significant reduction in total volume, fill up with desalted water and boil again briefly.

Gel does not polymerise completely

- The agarose wasn't dissolved / boiled thoroughly enough or the temperature during polymerisation was too warm. Allow the gel to harden for 10 minutes in the refrigerator.

^{*}compatible with multi-channel pipettors

Gel doesn't run / no air bubbles on the electrodes

- Check all connections, plugs and switches. Ensure that the level of the buffer is just above the top side of the gel.

Samples do not run cleanly out of the wells

- The wells were damaged. Pull the combs out even more carefully and cover the gel beforehand with a thin layer of running buffer.
- The bottom of the wells is damaged. Insert the comb in such a way that there is more space between the comb and the bottom of the gel casting tray.
- The DNA-samples contained debris.

Air bubbles appear in the gel during the run

- The agarose was not degassed thoroughly enough. This is particularly important when working with highly concentrated gels. Please ensure that the agarose is boiled well before casting to degas it. In the event of a significant reduction in total volume, fill up with desalted water and boil again briefly.

Bands are not straight, the border of the run shows a curve

- The agarose was not polymerised evenly.
- The gel is uneven in height. Please ensure that the gel casting tray lies flush during polymerisation. Use the levelling table.

Vertical smears in bands

- The gel mix may have contained dirt particles. Using clean water, additionally rinse the glass vessels before preparing the agarose.
- Too much DNA was applied. Dilute the sample.

Bands are diffuse

- A very high voltage may reduce the running time, but this results in poorer separation of the DNA. Reduce the voltage during the run.
- Running capacity of buffer was exceeded. Use a fresh buffer and check the buffer stock solution.
- The agarose wasn't dissolved / boiled thoroughly enough or the temperature during polymerisation was too warm. Allow the gel to harden for 10 minutes in the refrigerator.

Geltray is deformed / electrodes are broken

- Excessive voltage has been applied
- -The gel casting damswere not removed before the run

Electrophoresis Unit ROTIPHORESE® PROfessional I	2788.1
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Electrophoresis Unit ROTIPHORESE® PROfessional III	2850.1
Electrophoresis Unit ROTIPHORESE® PROfessional III Stretch 20	9938.1
Electrophoresis Unit ROTIPHORESE® PROfessional III Stretch 25	9939.1
Electrophoresis Unit ROTIPHORESE® PROfessional IV	2941.1
Electrophoresis Unit ROTIPHORESE® PROfessional V	3000.1



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