Instructions for use

ROTIPHORESE® Ready-to-Use Gel Solutions

3029, 3030, A515, T802, A516, A121, A124

Hazard and precautionary statements see page 3 of this instructions for use.

A) SDS-PAGE

Resolving gel *(data apply to 20 ml gel solution)*

30 % acryl-amide mix	**Gel concentration**	**6 %**	**8 %**	**10 %**	**12 %**	**15 %**
	Aqua dest. (ml)	10.6	9.3	7.9	6.6	4.6
	30 % acrylamide mix (ml)	4	5.3	6.7	8	10
	Tris (1.5 M, pH 8.8) (ml)	5	5	5	5	5
40 % acryl-amide mix	**Gel concentration**	**6 %**	**8 %**	**10 %**	**12 %**	**15 %**
	Aqua dest. (ml)	11.6	10.6	9.6	8.6	7.1
	40 % acrylamide mix (ml)	3	4	5	6	7.5
	Tris (1.5 M, pH 8.8) (ml)	5	5	5	5	5

Add in this order:
200 µl 10 % SDS solution (mix carefully, avoid bubbles)
200 µl 10 % ammonia persulphate solution (prepare freshly)
20 µl TEMED (mix carefully, avoid bubbles)
Pour gel immediately and overlay with isopropanol

Stacking gel *(data apply to 5 % gels)*

30 % acryl-amide mix	**Gel volume**	**1 ml**	**3 ml**	**5 ml**	**8 ml**	**10 ml**
	Aqua dest. (ml)	0.68	2.1	3.4	5.5	6.8
	30 % acrylamide mix (ml)	0.17	0.5	0.83	1.3	1.7
	Tris (1.0 M, pH 6.8) (ml)	0.13	0.38	0.63	1	1.25
	SDS (10 % solution) (µl)	10	30	50	80	100
	APS (10 % solution*) (µl)	10	30	50	80	100
	TEMED (µl)	1	3	5	8	10

40 % acryl-amide mix	**Gel volume**	**1 ml**	**3 ml**	**5 ml**	**8 ml**	**10 ml**
	Aqua dest. (ml)	0.725	2.185	3.645	5.84	6.3
	40 % acrylamide mix (ml)	0.125	0.375	0.625	1	1.25
	Tris (1.0 M, pH 6.8) (ml)	0.13	0.38	0.63	1	1.25
	SDS (10 % solution) (µl)	10	30	50	80	100
	APS (10 % solution*) (µl)	10	30	50	80	100
	TEMED (µl)	1	3	5	8	10

* prepare freshly!

Be careful to mix the solution thoroughly before and after addition of SDS and TEMED.
Avoid bubbles. Pour the stacking gel immediately and insert the comb carefully.

B) Separation of nucleic acids

Denaturing TBE gels for separation of single stranded nucleic acids (e.g. sequencing gels)
(data apply to 100 ml gel solution)

25 % sequencing gel concentrate with urea	**Gel concentration**	**4 %**	**6 %**	**8 %**
	Sequencing gel diluent (ml)*	74	66	58
	25 % sequencing gel concentrate (ml)	16	24	32
30 % acrylamide mix (29:1)	**Gel concentration**	**4 %**	**6 %**	**8 %**
	Aqua dest. (ad 90 ml) (ml)*	app. 52	app. 45	app. 39
	30 % acrylamide mix (ml)	13.3	20	26.5
	Urea (g)**	42	42	42
40 % acrylamide mix (19:1 or 29:1)	**Gel concentration**	**4 %**	**6 %**	**8 %**
	Aqua dest. (ad 90 ml) (ml)*	app. 55	app. 50	app. 45
	40 % acrylamide mix (ml)	10	15	20
	Urea (g)**	42	42	42

*If required for resolution of secondary structures the gel may be supplemented with formaldehyde by replacing 25 ml aqua dest. with 25 ml formaldehyde.
**Results in gels with 42 % urea (7 M)

Add in this order:
10 ml 10 x TBE buffer*** (mix carefully, avoid bubbles, degas if required)
400 µl 10 % ammonia persulphate solution (prepare freshly)
50 µl TEMED (mix carefully, avoid bubbles)
Pour gel immediately and insert the comb carefully

***Results in 45 % urea if sequencing gel concentrate and sequencing gel diluent are used. If 50% urea are required replace 10 x TBE by the ready-to-use sequencing gel buffer concentrate with 50 % urea (Ord. No. 3050.1).

TBE gels for electrophoresis of ds nucleic acid
(data apply to 100 ml gel solution)

30 % acrylamide mix (29:1)	**Gel concentration**	**6 %**	**10 %**	**15 %**
	Aqua dest. (ml)	69	56	39
	30 % acrylamide mix (ml)	20	33	50
40 % acrylamide mix (19:1 or 29:1)	**Gel concentration**	**6 %**	**10 %**	**15 %**
	Aqua dest. (ml)	74	64	51.5
	40 % acrylamide mix (ml)	15	25	37.5

Add in this order:
10 ml 10 x TBE buffer (mix carefully, avoid bubbles, degas if required)
1 ml 10 % ammonia persulphate solution (prepare freshly)
60 µl TEMED (mix carefully, avoid bubbles)
Pour gel immediately and insert the comb carefully

Detailed instructions for use and further information can be found in the info brochure PAGE Instructions (see www.carlroth.de next to the product description).

Hazard and precautionary statements

Please wear protective gloves and eye protection when handling the solutions.

Waste (solution residues, poured gels) must be disposed of as hazardous waste in accordance with the relevant national or regional provisions.

Please note safety data given on label and MSDS.

- **ROTIPHORESE® Ready-to-Use Gel Solutions: 3029, A124**

Danger H302-H315-H319-H317-H340-H350-H361f-H372
P201-P280-P301+P312-P302+P352-P305+P351+P338-P308+P313

- **ROTIPHORESE® Ready-to-Use Gel Solutions: 3030, A516, A121, A515, T802**

Danger H301-H315-H319-H317-H332-H340-H350-H361f-H372
P201-P280-P301+P310-P302+P352-P305+P351+P338-P308+P313

Art. No.	Product	Conc.	Mixing ratio
Acrylamide/bisacrylamide mixtures, ready-to-use			
3029	ROTIPHORESE® Gel 30 (37.5:1):	30 %	37.5:1
3030	ROTIPHORESE® Gel 40 (19:1):	40 %	19:1
A515	ROTIPHORESE® Gel 40 (29:1):	40 %	29:1
T802	ROTIPHORESE® Gel 40 (37.5:1):	40 %	37.5:1
Acrylamide/bisacrylamide mixtures for automated sequencing (fluorescence free)			
A516	ROTIPHORESE® NF acrylamide/bis-solution	40 %	19:1
A121	ROTIPHORESE® NF acrylamide/bis-solution	40 %	29:1
A124	ROTIPHORESE® NF acrylamide/bis-solution	30 %	29:1

Further ROTIPHORESE® Gel Solutions

Art. No.	Product	Conc.	Mixing ratio
Acrylamide- and bisacrylamide solution, ready-to-mix			
3037	ROTIPHORESE® Gel A acrylamide solution	30 %	x
7748	ROTIPHORESE® Gel A-40 acrylamide solution	40 %	x
3039	ROTIPHORESE® Gel B bisacrylamide solution	2 %	x
Sequencing gel solutions, ready-to-use			
A431	ROTIPHORESE® DNA sequencing system	Kit	3043. 3047. 3050
3043	ROTIPHORESE® sequencing gel concentrate: urea and acrylamide/bisacrylamide	50 % 25 %	 19:1
3047	ROTIPHORESE® sequencing gel diluent: urea	50 %	
3050	ROTIPHORESE® sequencing gel buffer concentrate urea in 10x TBE	 50 %.	

ROTIPHORESE®Gel 30 (37,5:1)	**glass**	**1 L**	**3029**
ROTIPHORESE®Gel 40 (19:1)	**glass**	**1 L**	**3030**
ROTIPHORESE®Gel 40 (29:1)	**glass**	**1 L**	**A515**
ROTIPHORESE®Gel 40 (37,5:1)	**glass**	**1 L**	**T802**
ROTIPHORESE®NF-Acrylamid/Bis-solution 40 %(19:1)	**glass**	**250 ml**	**A516**
ROTIPHORESE®NF-Acrylamid/Bis-solution 40 % (29:1)	**glass**	**250 ml**	**A121**
ROTIPHORESE®NF-Acrylamid/Bis-solution 30 % (29:1)	**glass**	**1 L**	**A124**

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