# **InterBioTech**



Crosslinking agent for the preparation of gel electrophoresis (polymerization of acrylamide).

# **Product Description**

Crosslinkers S	Selection	Guide	
Name	Cat.#	Chemical Structure	Benefits
Bis- Acrylamide	05379L	$H_2C \longrightarrow N \longrightarrow N \longrightarrow CH_2$	General crosslinker in PAGElectrophoresis
PDA	1A5041		Reduce silver stain background in SDS- PAGE and 2-D gels, increases resolution, et gives higher gel strength
DATD	118221	$H_2C \xrightarrow{H} N \xrightarrow{OH} N \xrightarrow{OH} H$	Increase pore size of IEF gels, where molecular sieving is a problem. Used in scintillation counting: 1,2-dio structure is soluble in periodic acid
Name:	Bis-A	crylamide (bis)	<b>Catalog #:</b> 05379L, 100g powder
Storage:	N,N'-me EC [20 +4°C (	thylene-bis-acrylamide 3-750-9]; CAS: [110-26-9]; MW: 154.17. or –20°C for long term) (K)	UP864965, solution 2% Standard crosslinking agent for the preparation of polyacrylamide gel electrophoresis
Name:	N-N'	diallyltartardiamide (DATD)	Catalog #: 118221, 1g
Storage:	N-N' dia EC : 20 +4°C (	llyl-tartardiamide 61-277-3; CAS: 28843-34-7; MW: 228.25 or –20°C for long term) (K)	A usefull crosslinking agent for the preparation of polyacrylamide gel electrophoresis
Name:	Piper	azine diacrylamide (PDA)	Catalog #: 1A5041, 1g
	1,4-Bis(acryloyl)piperazine EC : 261-277-3; CAS: 58477-85-3; MW: 194.23 Light yellow powder Fully miscible in water. A usefull crosslinking a polyacrylamide gel elec Kelkar RS, Mahen AA, Saoji AM as a cross-linking agent for polyac		A usefull crosslinking agent for the preparation of polyacrylamide gel electrophoresis Kelkar RS, Mahen AA, Saoji AM, Kelkar SS. N-N' diallyltartardiamide (DATE as a cross-linking agent for polyacrylamide gel disc electrophoresis of human
Storage:	+4°C (	or $-20^{\circ}$ C for long term) (K)	serum proteins. J Postgrad Med [serial online] 1986 [cited 2013 Feb 14];32:27- 31. <u>Article</u>
Also available	: N,N'- CAS:	(1,2-Dihydroxyethylene)bisacrylamide #Bl 868-63-3; EC:212-280-1	$H_{2}C \xrightarrow{H} O \xrightarrow{H} O \xrightarrow{H} O \xrightarrow{H} O \xrightarrow{CH_{2}} CH_{2}$

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## FT-1A5041 PDA information

#### Introduction <sup>[r]</sup>

The PDA crosslinking agent can replace methylene-bis-acrylamide (bis), the standard crosslinker used to prepare polyacrylamide gels for PAGE, IEF, and protein sequencing. Benefits include:

- particularly reduced background for silver staining,
- increased gel strength, and
- higher resolution gels.

Standard polymerization protocols used for bis work for PDA.

PDA was designed to reduce the background; in consideration that basic and sulfur-containing amino acids are essential in the detection of peptides by the silver stain reaction, the piperazine ring of PDA eliminates the hydrogen atoms of the amide groups. Correlately, PDA improves the signal to noise and thus silver staining sensitivity. It also gives benefits in polyacrylamide electrophoresis gels:

<u>Application</u>	Advantages of substituting bi by PDA	
SDS-PAGE	• Increased gel strength with low %T gels	
	Reduced background in silver staining	
2-D gels	Increased gel strength facilitates tube gel handling	
	• Tube gels are more stable (can be stored for 1 month at 4 °C without precipitation of urea).	
	Reduced background in silver staining	
	Increased resolution of protein spots	
Protein sequencing gels	• Decreased N-terminal blockage increases sensitivity of micro sequencing from 2-D blots	

To start using PDA, simply substitute PDA gram for gram for bis in your usual procedure, keeping current polymerization conditions and catalyst concentrations used for bis-acrylamide (see <u>FT-86489B</u>).

#### <u>Usage</u>

Usually the pore size of the polyacrylamide gel is changed by adjusting the total monomer concentration  $(\%T)^{[note]}$ : the monomer / polymer solution is prepared with constant monomer relative concentration %C, and its volume is adjusted depending on desired pore size.

%T= [(g Acrylamide + g PDA) / Total Volume] x 100

%C= [g PDA/(g Acrylamide + g PDA)] x 100

With PDA concentration increasing from 2 to 5%, the pore size of the gel decreases and the protein mobility decreases (higher concentrations is not recommended, because it turns the gel opaque and increases the mobility of proteins).

#### Standard formulation of separating gel and stacking gel solutions:

#### • Acrylamide/PDA stock solution (30% T, 2.67% C):

dissolve 4.0 g of PDA and 146.0 g of acrylamide to 500 ml with distilled water. Fillter and store in the dark at 4 °C.

• Separating gels (0.375M Tris, pH 8.8, with <u>7.5% PDA</u>):

add 25ml of Acrylamide/PDA solution (30% T, 2.67% C) and complete to 73.5ml with distilled water. Add 25ml of 1.5M Tris HCl pH8.8, 1ml of SDS 10%(w/v), 500µL of APS 10% (fresh) and 50µL of TEMED.

<i>Note</i> : The volume of Acrylamide/P	DA stock required for a desired total monomer concentration (X) can be calculated
as follows:	volume $(30\% \text{ T}, 2.67\% \text{ C stock soln}) = (X \% \text{T}) \text{ x} (3.33)$ .

Then complete with water: volume of water to add = 73.5ml - (volume used of 30% T, 2.67% C stock).

*Note*: For optimal results, leave the acrylamide/PDA mix solution under vacuum for at least 15 minutes before adding the two catalysts, just prior to casting the gels.

#### • Stacking gel (0.125M Tris pH 6.8 <u>4% PDA</u>): ,

add 1.3ml of PDA solution (2.67% C) and complete to 7.35ml with distilled water. Add 100µl of Tris 0.5M Tris HCl pH6.8, 50µl of SDS 10%(w/v), 50µlof APS 10% (fresh) and 10µL of TEMED.



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*Note*: a higher TEMED concentration and faster polymerization are required for the stacking gel because of the inhibitory effect of atmospheric oxygen associated with the comb.

#### • Gel storage

*Remark*: PDA crosslinked gels shrink more than bis crosslinked gels when they are dried after equilibration in solutions containing only alcohol and acetic acid. Addition of 3-5% glycerol to the equilibration solution should overcome any shrinking problems.

#### Legals:

• For Research Use Only

• Safety (Regulation (EC) No 1272/2008)

Symbol: GHS07

Skin Irrit. 2 H315 Causes skin irritation.

Eye Irrit. 2 H319 Causes serious eye irritation.

STOT SE 3 H335 May cause respiratory irritation.

P261 Avoid breathing dust/fume/gas/mist/vapours/spray.

P280 Wear protective gloves/protective clothing/eye protection/face protection.

P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. P321 Specific treatment (see on this label).

P405 Store locked up.

P501 Dispose of contents/container in accordance with local/regional/national/international regulations

• Safety(Classification according to Directive 67/548/EEC or Directive 1999/45/EC) Xi; Irritant R36/37/38: Irritating to eyes, respiratory system and skin.

#### Related / associated products and documents

Ammonium Persulfate (APS) UP306098TEMED UP15413DUrea UP031903Acrylamide 40% solutions FT-86489BOther electrophoresis reagentssee Product hightlights, BioSciences Innovations catalogue and e-search tool.

## **Ordering information**

Catalog size quantities and prices may be found at <u>http://www.interchim.com</u>. Please inquire for higher quantities (availability, shipment conditions).

Please contact InterBioTech – Interchim for any other information Hotline : +33(0)4 70 03 73 06 – <u>Interbiotech@interchim.com</u>

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