

Formamide deionized Molecular biology grade

Formic acid amide

Product No. A2156

Description

Formula:	CH ₃ NO
Molecular weight:	45.04 g/mol
CAS-No.:	[75-12-7]
HS-No.:	29241000
Assay (from N):	min. 99.5 %
Refractive index (n 20°/D):	1.4472
Melting point:	2°C
Boiling point:	210°C
Working concentration:	10 - 100 mM
Storage:	Room temperature
Safety:	R: 61 / WGK 1 S: 53-24/25-37-45 toxic for reproduction
Disposal:	1

Specification:	
DNases/RNases/Proteases	not detectable
Assay (from N)	min. 99.5 %
Fe	max. 0.00001 %
Water	max. 0.1 %
Pb	max. 0.00001 %
Chloride	max. 0.00005 %

Comment

RNA and **DNA** with a chain length of >150 - 200 base pairs completely denature at room temperature in 98 % formamide, but not in 7 M urea. This allows the exact determination of the size of DNA or RNA single strands, because under these conditions the base composition and secondary structure have no influence on the migration behavior. The polyacrylamide gel contains 98 % deionized, anhydrous formamide. Acrylamide and bisacrylamide are dissolved in the formamide (1-4). The loading buffer contains formamide, too (see AppliChem's loading buffers!). A similar technique of formamide-containing sequencing gels can be used for the sequencing of nucleic acids, when the secondary structure of the sequencing product causes an anomalous migration, i.e. compression of bands. Inclusion of up to 40 % of formamide is possible to overcome this problem (Ref. 8, pages 7.6.1-7.6.10).

The stability of RNA is in formamide higher than in DEPC-treated water. RNA may be stored for more than a year in formamide at -20°C, instead of storage at -70°C in DEPC-treated water.

Besides gel electrophoresis deionized formamide is used for the hybridization of nucleic acids (Ref. 8, pages 2.10.1-2.10.16 + 6.3.1-6.3.3). Formamide reduces the melting temperature of a DNA-DNA hybrid by an average of 0.6°C per 1 % formamide; the maximum concentration is 50 %. By adding formamide, the temperature during the hybridization process can be reduced in comparison to an aqueous solution. At lower temperatures less DNA will detach from the nitrocellulose membrane. In addition, the background hybridization of heterologues RNA probes will be reduced. In combination with nylon membranes, no major advantage was observed.

A typical formamide-containing prehybridization/hybridization solution may be composed of: 5X SSC, 5X Denhardt solution, 50 % (w/v) formamide, 1 % (w/v) SDS and 100 µg/ml denatured salmon sperm DNA added just before use (ref. 8, p. 2.10.7). The use of formamide-containing buffers increase the specific hybridisation in blotting experiments and reduces thereby the number of washing steps. This should be taken into account, if one switches to buffers without formamide (6).

Application and Literature

- (1)Pinder, J.C.*et al.* Properties of RNA in Formamide. (1974) *Biochemistry* **13**, 5367-5373
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- (3)Maniatis, T. & Efstratiadis, A. Fractionation of low molecular weight DNA or RNA on polyacrylamide gels containing 98 % Formamide or 7 M Urea. (1980) *Methods Enzymol.* **65**, 299-305
- (4)Meinkoth, J. & Wahl. G. Hybridisation of nucleic acids, immobilized to membranes. (1984) *Anal. Biochem.* **138**, 267-284
- (5)Chomczynski, P. Storage of RNA in formamide increases stability of the RNA. (1992) *Nucleic Acids Res.* **20**, 3791-3792
- (6)Cornish, E.C. *et al.* Improvement of the Southern blot method: Influence of the hybridisation buffer. (1998) *BioTechniques* **25**, 948-954
- (7)Kafatos, F.C. *et al.* Determination of nucleic acid sequence homologies with 'dot hybridization'. (1979) *Nucleic Acids Res.* **7**, 1541-1552
- (8)Ausubel, F.A., Brent, R., Kingston, R.E., Moore, D.D., Seidman, J.G., Smith, J.A. & Struhl, K. (eds.) 2001. *Current Protocols in Molecular Biology*. John Wiley & Sons, N.Y.