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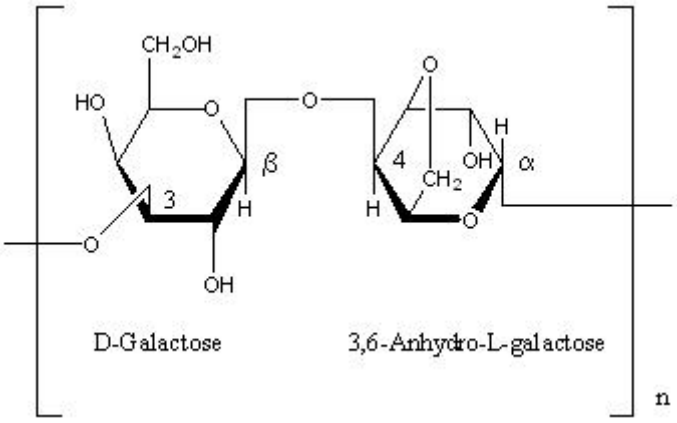
TECHNICAL INFORMATION

Catalog Number: 193983, 193984, 193985, 193986, 193987, 800257, 800259, 800568, 800666, 800667, 800668, 800669, 800670, 800671, 820721, 820723, 952012, 952014, 952015, 952017

Agarose

Structure:

Agarose is a linear polymer consisting of alternating D-galactose and 3,6-anhydro-L-galactose units.



CAS #: 9012-36-6

Synonyms: 3,6-Anhydro-a-L-galacto-b-D-galactan; Sepharose

Physical Properties:

Purity: Sulfate Content is usually an indicator of purity. Sulfate is the major ionic group present as an impurity in agarose (see certificate of analysis for individual lots).

Gel Strength: This is the force that must be applied to a gel to cause it to fracture.

Gel Point: The Temperature at which an aqueous agarose solution forms a gel. The gel point is not the same as melting point of agarose because of solutions exhibiting hysteresis in the liquid to gel transition.

Electroendosmosis (EEO): A movement of liquid through the gel. Anionic groups in an agarose gel are affixed to the matrix and cannot move, but dissociable cations can migrate toward the cathode in the electrophoresis unit. Since electrophoretic movement of biopolymers is usually toward the anode, EEO can disrupt separations because of internal convection.

Description: Agarose is a purified linear galactan hydrocolloid isolated from agar or agar-bearing marine algae.

Some Typical General Uses: Used to separate nucleic acids electrophoretically because agarose gels have larger pore sizes than cross linked acrylamide gels at low concentrations. Agarose gels tend to be more solid, but less elastic, than cross-linked acrylamide gels. Agarose gels are also used in immunoelectrophoresis (IEP) and double-diffusion Ouchterlony plates to demonstrate antibody-antigen reactions. Used in chromatographic separations by either beading the agarose or cross-linking it.[1]

Typical Methods for Gelation:

Boiling water bath method:

– Add any buffer or choice (usually with an ionic strength, u,[2] of 0.03-0.10) and a stir bar to a beaker which can hold 2-4 times

the volume of the desired solution.[3]
– Slowly sprinkle the agarose powder into the liquid while stirring to prevent clumping.
– Weigh the beaker and solution before heating.
– Cover the beaker with plastic wrap and pierce a hole in the wrap for ventilation.
– Bring the solution to a boil and allow it to boil for 5-10 minutes stirring continuously, until agarose dissolves completely. To avoid charring, use a boiling water bath rather than directly applied heat.[3]
– Add enough hot distilled water to return the contents to the original weight; mix continuously.
– Allow the mixture to cool to 50-55°C, at which temperature it is ready to be cast into cassettes which have been pre-warmed to 50-55°C.

Microwave Method 1 (for gels not more than 2% w/v):

– Add any buffer of choice (usually with an ionic strength, u,[2] of 0.03-0.10) and a stir bar to a beaker which can hold 2-4 times the volume of the desired solution.[3]
– Slowly sprinkle the agarose powder into the liquid while stirring to prevent clumping.
– Remove the stir bar.
– Weigh the beaker and solution before heating.
– Cover the beaker with plastic wrap and pierce a hole in the wrap for ventilation.
– Place the solution in a microwave oven and heat on HIGH power for 2 minutes.
– Remove the solution from the oven very carefully; any microwaved solution may be superheated and could foam over the container's rim if agitated. Swirl gently to re-suspend any remaining agarose particles.
– Reheat on HIGH power for 1-2 minutes or until the solution comes to a boil. Boil for 1 more minute or until the solution is clear and the agarose is completely dissolved.
– Remove the solution from the oven very carefully and swirl it gently.
– Add enough hot distilled water to return the contents to the original weight; mix continuously.
– Allow the mixture to cool to 50-55°C, at which temperature it is ready to be cast into cassettes which have been prewarmed to 50-55°C.[3]

Microwave Method 2 (for gels greater than 2% w/v):

Follow the same protocol as Microwave Method 1, but use a MEDIUM instead of HIGH power setting in step 6.

Agarose Gels for Double-Diffusion Assays (Ouchterlony):

Follow either the boiling method or Microwave method 1. Use an agarose concentration of approximately 0.9% w/v. For the buffer use a borate saline buffer, pH 8.5, ionic strength 0.175, with 0.01% thimerosal and 3% polyethylene glycol (PEG). PEG enhances the formation of precipitin, increasing assay sensitivity. Trypan blue can also be added as an indicator. Trypan blue improves visualization of precipitin lines by direct light, when a "dark field" viewer is not available.

Availability:

Catalog Number	Description	Gel Strength (gm/cm^2)	Gel Temp.	EEO	Sizes
820721 820723	Agarose, Electrophoresis Grade	≥ 600	36°C	0.13	100 g 500 g
952014 952012 952015 952017	Agarose, Immunodiffusion Grade	Approx. 2000	36°C	0.12	50 g 250 g 1 kg 5 kg
800668 800568 800669	Agarose, Genetic Technology Grade	≥ 2000	36°C	0.17	100 g 125 g 500 g
800666 800667	Agarose, High Gel Temperature	≥ 1200	42°C	0.1	25 g 100 g
800670	Agarose, For Isoelectric Focusing	≥ 700	41-43°C	0.00	10 g 25 g
800257 800259	Agarose, Low Gel Temperature	≥ 550	< 30°C	0.1	25 g 100 g
193983	Agarose, Molecular Biology Reagent	≥ 600	36°C	0.09-0.13	10 g 25 g 50 g 100 g 250 g 500 g

193984	Agarose, High Resolution, Molecular Biology Reagent; Separates small DNA fragments (200-800 bp) with a resolution comparable to acrylamide	≥ 600	< 30°C; A 1% solution remains fluid at 37°C for up to 24 hours. Will to a firm gel at < 25°C, and not remelt until temperatures exceed 65°C. This ability to remain in solution at 30-37°C allows a second digest on a restriction enzyme fragment without need to recover it from the gel.	≤ 0.12	25 g 100 g
193985	Agarose, Pulsed Field Electrophoresis, Molecular Biology Reagent; Ideal for high molecular weight DNA separation	≥ 600	36°C	0.1 ± 0.02	25 g 100 g 250 g
193986	Agarose, Pulsed Field Electrophoresis Sample Preparation, Molecular Biology Reagent; For making gel plugs for high molecular weight DNA separation.	≥ 600	< 30°C	0.1	500 mg 1 g
193987	Agarose, Broad Range, Molecular Biology Reagent; This is a general use agarose capable of separating DNA from 50 to 1000 bp on a 3% gel.	Approx. 600	≤ 35°C	< 0.15	5 g 10 g 25 g 100 g 500 g

References:

– *Gel Filtration Principles and Methods, 5th Ed.*, Pharmacia LKB Biotechnology (1991).
– $u = 2S_iC_ixZ_i^2$

where:C_i = molar concentration of a given ion
Z_i = charge of a given ion

Cooper, T.G., *The Tools of Biochemistry*, John Wiley & Sons, New York, p. 176 (1977).
3. Andrews, A.T., *Electrophoresis: Theory, Techniques, and Biochemical and Clinical Applications, 2nd Ed.*, Peacock, A.R. and Harringdon, W.F. (eds.), Oxford Science Publications, Clarendon Press, Oxford, p. 149 (1993).