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# **ProductInformation**

#### **AGAROSE**

**CAS NUMBER:** 9012-36-6

CAS NUMBER: 39346-81-1 (products A9414,

A4018, A6560, A9045, A0701)

**SYNONYMS:** 3,6-Anhydro-α-L-galacto-β-D-galactan; FastLane agarose; Indubiose A4; NuSieve GTG; Odigose;

Seakem; Sepharose

#### **PRODUCT DESCRIPTION - APPLICATIONS:**

Agarose is a purified linear galactan hydrocolloid isolated from agar or agar-bearing marine algae. Structurally, it is a linear polymer consisting of alternating D-galactose and 3,6-anhydro-L-galactose units. As a gelling agent, agarose is used: 1.) to separate nucleic acids electrophoretically because its gels have larger pore sizes than polyacrylamide gels at low concentrations. Unlike polyacrylamide, the consistency of the gels is more solid (but also less elastic); 2.) To demonstrate cross reaction in IEP (Immuno electrophoresis) and Ouchterlony (double diffusion) plates in which antibody-antigen precipitin lines are studied; 3.) to make gel plates or overlays for cells in tissue culture. 4.) To form a gel matrix (either beaded and/or crosslinked) which can be used in chromatographic separations.<sup>1</sup>

#### PROPERTIES OF AGAROSES OFFERED BY SIGMA:

Please refer to the table on pages 4-8.

Sulfate content may be used as an indicator of purity, since sulfate is the major ionic group present.

**Gel strength** is the force that must be applied to a gel to cause it to fracture.

The **gel point** is the temperature at which an aqueous agarose solution forms a gel as it cools. Agarose solutions exhibit hysteresis in the liquid-to-gel transition - that is, their gel point is not the same as their melting temperature.

#### PROPERTIES OF AGAROSES OFFERED BY SIGMA: (continued)

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, giving rise to **electroendosmosis (EEO)** - a movement of liquid through the gel. Since electrophoretic movement of biopolymers is usually toward the anode, EEO can disrupt separations because of internal convection.

#### **USAGE INSTRUCTIONS FOR MAKING GELS:**

#### **Boiling water bath method:**

- a. Add any buffer of choice (usually with an ionic strength,  $\mu$ , of 0.03-0.10) and a stir bar to a beaker which can hold 2-4 times the volume of the desired solution.
- b. Slowly sprinkle the agarose powder into the liquid while stirring to prevent clumping.
- c. Weigh the beaker and solution before heating.
- d. Cover the beaker with plastic wrap and pierce a hole in the wrap for ventilation.
- e. 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.<sup>3</sup>
- f. Add enough hot distilled water to return the contents to the original weight; mix continuously.
- g. 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.<sup>3</sup>

### Microwave method 1 (for gels £2% w/v):

- a. Add any buffer of choice (usually with an ionic strength,  $\mu$ , of 0.03-0.10) and a stir bar to a beaker which can hold 2-4 times the volume of the desired solution.
- b. Slowly sprinkle the agarose powder into the liquid while stirring to prevent clumping.
- c. Remove the stir bar.
- d. Weigh the beaker and solution before heating.
- e. Cover the beaker with plastic wrap and pierce a hole in the wrap for ventilation.
- f. Place the solution in a microwave oven and heat on HIGH power for 2 minutes.
- g. 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.
- h. 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.
- i. 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.
- k. 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.<sup>3</sup>

# Microwave method 2 (for gels >2% w/v):

Follow the same protocol as for ≤2% gels (above), but use a MEDIUM instead of HIGH power setting in step f.

#### OTHER GELLING AGENTS:

- 1. Agar: Separate data sheet is available.
- 2. Gelatin: Separate data sheet is available.
- 3. Phytagel (Sigma product number P8169): Phytagel is an agar substitute produced from a bacterial carbohydrate composed of glucuronic acid, rhamnose and glucose. It produces a clear, colorless, high strength gel which aids in detection of microbial contamination. Phytagel provides an economical alternative to agar as a gelling agent (agar contain about 70% agarose). Originally developed for microbial applications, Phytagel is a good choice in any application (such as plant cell culture) where some agar products tend to inhibit growth due to unidentified impurities. To prevent clumping, Phytagel should be added to culture medium that is at room temperature, with rapid stirring.
- 4. Agargel (Sigma product number A3301): Agargel is a blend of agar and Phytagel which was developed to help control vitrification in plant tissue cultures. Agargel provides the positive attributes of agar and Phytagel, is cheaper than agar, and is superior to Phytagel in applications where vitrification is a problem. Agargel produces a semiclear gel which allows for better detection of contamination.

# **REFERENCES:**

- 1. Gel Filtration Principles and Methods, 5th Ed., Pharmacia LKB Biotechnology (1991).
- 2.  $\mu = 2 S_i C_i \times Z_i^2$

 $C_i$  = molar concentration of a given ion

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

PRODUCT	NAME - DESCRIPTION	SULFATE	GEL STRENGTH (g/cm²)	GEL POINT (°C)	MELTING T. (°C)	EEO
A0169	Agarose Type I-A: Low EEO	<0.20%	>1200 at 1.0% >2500 at 1.5%	36±1.5 at 1.5%	87±1.5	0.09-0.13
A0576	Agarose Type I-B: Low EEO Exceptionally high gel strength makes this agarose particularly suitable for separating high molecular weight nucleic acids at low gel concentrations	≤0.12%	≥1800 at 1.0% ≥3200 at 1.5%	36±1.5 at 1.5%	86±2.0	≤0.12
A6013	Agarose Type I: Low EEO	≤0.15%	≥1200 at 1.0%	36±1.5 at 1.5%	N/A	0.09-0.13
A9918	Agarose Type II-A: Medium EEO	<0.25%	>1000 at 1.0%	36±1.5 at 1.5%	87±1.5	0.16-0.19
A6877	Agarose Type II: Medium EEO	≤0.20%	≥1000 at 1.0%	36±1.5 at 1.5%	N/A	0.16-0.19
A9793	Agarose Type III-A: High EEO	<0.25%	>750 at 1.0% >1000 at 1.5%	36±1.5 at 1.5%	87±1.5	0.23-0.26
A6138	Agarose Type III: High EEO	≤0.20%	≥650 at 1.0%	36±1.5 at 1.5%	N/A	0.23-0.26
A9668	Agarose Type IV-A: Special High EEO	<0.30%	>700 at 1.0% >1000 at 1.5%	35±1.5 at 1.5%	87±1.5	≥0.30
A3643	Agarose Type IV: Special High EEO Characterized by lower sulfate content and lower non-specific protein binding capacity than Type III. It is useful for electrophoretic techniques requiring a high degree of cathodal movement. Trailing and smearing of protein due to non-specific binding is minimized.	≤0.25%	≥650 at 1.0%	36±1.5 at 1.5%	N/A	≥0.30
A3768	Agarose Type V: High Gelling Temperature Higher gelling Temperature and lower EEO the Type I	≤0.30%	≥800 at 1.0%	42±1.5 at 1.5%	N/A	≤0.10
A7174	Agarose Type VI-A: High Gelling Temperature	<0.20%	>900 at 1.0% >1200 at 1.5%	41±1.5 at 1.5%	95±1.5	≤0.14
A3893	Agarose Type VI: High Gelling Temperature Higher gel strength than type V	≤0.20%	≥1000 at 1.5%	42±1.5 at 1.5%	N/A	<0.10

PRODUCT	NAME - DESCRIPTION	SULFATE	GEL STRENGTH (g/cm²)	GEL POINT (°C)	MELTING T. (°C)	EEO
A0701	Agarose Type VII-A: Low Gelling Temperature Excellent for in-gel enzymatic reactions and cloning assay and for recovery of heat-labile samples after electrophoresis.	≤0.10%	≥250 at 1.0% 500 at 1.5%	26±2.0 at 1.5%	≤65.5	≤0.12
A4018	Agarose Type VII: Glow Gelling Temperature A low gelling temperature derivative with unique gelling properties. Gels form at <30EC and remelt at temperatures >65EC. Gels exhibit excellent clarity and are particularly useful for preparation of media containing heat-labile materials.	≤0.10%	≥200 at 1.0%	26-30 at 1.5%	≤65	≤0.10
A4905	Agarose Type VIII For isoelectric focusing. High gel strength. EEO not detectable.	≤0.20%	≥500 at 1.0%	N/D	N/A	N/D
A2576	Agarose Type IX-A: Ultra-low Gelling Temperature Yields unusually strong gels for an ultra-low gelling agarose. Ideal for electrophoresis of heat-labile samples and for growth of hybridomas and other cell lines.	≤0.14%	≥100 at 1.0% ≥400 at 1.5%	≤17 at 1.5%	≤60	≤0.11
A5030	Agarose Type IX: Ultra-low Gelling Temperature Gelling Temperature Gelling occurs at 8-17EC and remelt at <50EC	≤0.10%	≥75 at 2.0%	8-17 at 0.8%	≤50	≤0.05
A3038	Agarose Type XI: Low Gelling Temperature Suitable for separation of small nucleic acid fragments.	≤0.15%	≥500 at 4.0%	≤35 at 4.0%	≤65	≤0.15
A7299	Agarose Type XII: Low Viscosity for Beading Recommended for preparation of agarose beads.	≤0.20%	≥500 at 1.0% ≥900 at 1.5%	41±1.5 at 1.5%	87±1.5	<0.14

PRODUCT	NAME - DESCRIPTION	SULFATE	GEL STRENGTH (g/cm²)	GEL POINT (°C)	MELTING T.	EEO
A4679	Agarose  Electrophoresis Reagent  Suitable for standard immunoelectrophoresis and immunodiffusion	≤0.20%	≥1200 at 1.0%	36±1.5 at 1.5%	88±1.5	0.09-0.13
A9311	Agarose: Medium EEO Electrophoresis Reagent	≤0.35%	≥1000 at 1.0%	36±1.5 at 1.5%	N/A	0.16-0.19
A4804	Agarose  Electrophoresis Reagent  Suitable for isoelectric focusing.	≤0.10%	>700 at 1.5%	32±2.0 at 0.8%	N/A	<0.02
A5304	Agarose  Electrophoresis Reagent  Suitable for counterimmunoelectrophoresis and immunoelectrophoretic techniques with significant cathodal migration.	≤0.30%	≥700 at 1.0%	36±1.5 at 1.0%	N/A	≥0.30
A8455	Agarose  Molecular Biology Reagent  DNase and RNase: none detected.  Separates small DNA fragments (200-800 base pairs) with a resolution approximately equal to acrylamide.	N/A	N/A	<30	<70	#0.35
A6560	Agarose Type VII: Low Gelling Temperature  Plant Cell Culture Tested  A low gelling temperature derivative with unique gelling properties. Gels form at <30EC and remelt at >65EC. Gels exhibit excellent clarity and are useful for the preparation of media containing heat-labile materials.	≤0.10%	≥200 at 1.0%	26-30 at 1.5%	≤65	≤0.10
A9539	Agarose: For Routine Use  Molecular Biology Reagent  DNAse, RNase and protease: none detected.  Used routinely at Sigma for analysis and purification of nucleic acids	≤0.15%	≥1200 at 1.0%	36±1.5 at 1.5%	N/A	0.09-0.13

PRODUCT	NAME - DESCRIPTION	SULFATE	GEL STRENGTH (g/cm²)	GEL POINT (°C)	MELTING T.	EEO
A2929	Agarose: For Pulsed Field Electrophoresis Running Gel Molecular Biology Reagent DNase and RNase: none detected. Suitable for the separation of high molecular weight DNA. Gels are easy to handle and give faster separation and better resolution of high molecular weight DNA by field inversion electrophoresis than product A9539.	≤0.20%	N/A	42±1.0	N/A	≤0.08
A3054	Agarose: For Pulsed Field Electrophoresis Sample Preparation Molecular Biology Reagent DNase and RNase: none detected. Suitable for making gel plugs when separating high molecular weight DNA without the shearing encountered by conventional isolation techniques. While embedded in plugs made from this agarose, cells can be lysed and the released DNA can be digested with restriction endonucleases.	≤0.15%	N/A	≤30	N/A	≤0.10
A9414	Agarose: Low Melting Point  Molecular Biology Reagent  DNase, RNase and protease: none detected.	<0.10%	N/A	~30	~65	≤0.10
A2790	Agarose: Wide Range  Molecular Biology Reagent  DNase, RNase and protease: none detected.  Capable of separating DNA fragments with 50-1,000 base pairs on a single 3% gel.	<0.35%	N/A	≤35	≤65	<0.15
A7431	Agarose: Wide Range/Standard 3:1 Ratio  Molecular Biology Reagent  DNase and RNase: none detected.  Composed of 3 pars low gelling temperature agarose and 1 part high gelling temperature agarose, this product is specially formulated to form strong, flexible gels for separation of small (#1 kb) PCR products, DNA and RNA.	≤0.15%	N/A	34±1.5	≤90	N/A

PRODUCT	NAME - DESCRIPTION	SULFATE	GEL STRENGTH (g/cm²)	GEL POINT(°C)	MELTING T.	EEO
A9702	Agarose Gel Mixture:  1% Agarose in TAE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					
A9827	Agarose Gel Mixture: 1.5% Agarose in TAE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					
A9952	Agarose Gel Mixture: 2% Agarose in TAE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					
A0203	Agarose Gel Mixture:  1% Agarose in TBE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					
A0328	Agarose Gel Mixture: 1.5% Agarose in TBE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					
A0453	Agarose Gel Mixture: 2% Agarose in TBE Buffer  Molecular Biology Reagent  DNase, RNase and protease: none detected.					

N/A = Not Available N/D = Not Detectable = Not Applicable

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