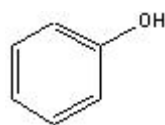


TECHNICAL INFORMATION

Catalog Number: 191423, 194011, 800672, 800673, 818048

Phenol

Structure:



Molecular Formula: C₆H₆O

Molecular Weight: 94.1

CAS # 108-95-2

Synonyms: Carboic acid; Phenic acid; Phenylic acid; Phenyl hydroxide; Hydrobenzene

Physical Description: White waxy solid. Product should not have any coloration such as pink to red. Note: Phenol is prone to redden on exposure to air and light, hastened by presence of alkalinity.¹ This oxidation makes it unsuitable for nucleic acid work. Oxidation products make DNA resistant to hydrolysis by DNase I. One such oxidation product is o-hydroxybiphenyl. Inhibition is due to the direct action on the DNA rather than on the enzyme, due to hydrogen bonding between bases of the nucleic acid.⁵

Solubility: Soluble in aqueous solution (5%-clear, colorless solution)

Extinction Coefficient: Em^M (ethanol) = 1.45 (276 nm), 1.91 (271 nm), 6.03 (218 nm)⁴

pKa: 10.0 @ 25°C¹

Description: Used as a general disinfectant, either in solution or mixed with slaked lime, etc., for toilets, stables, cesspools, floors, drains, etc.; for the manufacture of colorless or light-colored artificial resins, many medical and industrial organic compounds and dyes; as a reagent in chemical analysis.¹ Aqueous solutions are used as topical anesthetic; topical antiseptic; topical antipruritic.¹

Phenol is used in DNA extractions for removing protein. In an extraction for removing protein using phenol saturated with a buffer, at pH 8 or higher, DNA and RNA are more soluble in the upper, aqueous-rich layer leaving the proteins at the interface. At a pH below 7.0, the RNA remains soluble in the aqueous phase, but the DNA collects at the interface with the protein.

The DNA is removed from the aqueous layer with increasing efficiency as the pH is lowered with a maximum efficiency at pH 4.5. The phenol-rich phase does not separate well, so chloroform is added to force a cleaner separation of the organic and aqueous phases. Sometimes isoamyl alcohol is added as an antifoaming agent. A second extraction with just chloroform removes the residual phenol from the aqueous phase.

Protocol for the Preparation of Phenol : Chloroform: Isoamyl alcohol Solutions:^{2,3}

- Melt the phenol in a 45-50°C water bath.
- To a large beaker, add 1 volume of the melted phenol.
- Add [8-Hydroxyquinoline \(8-Quinolinol\)](#) to a concentration of 0.1% as a preservative. This is an optional step but will prolong the shelf life of the solution.
- To the melted phenol add an equal volume of [Tris-HCl buffer, pH 8.0](#) and stir with a magnetic stir bar for approximately 15 minutes. Stop stirring and allow the phases to separate. Carefully aspirate off the top aqueous layer. Repeat this step at least one more time and until the pH of the phenolic layer reaches a pH of > 7.8 as measured with pH paper or the below described pH method.
- After the final aqueous layer is removed, add 0.1 volume of [0.1 M Tris-HCl \(pH 8.\)](#). 0.2% [beta-mercaptoethanol](#) can be added

to the saturated (equilibrated) phenol solution if needed. This solution can then be stored in an amber bottle in the refrigerator for approximately 1 month.

– To make the phenol:chloroform:isoamyl alcohol solution, mix 25 volumes of the above equilibrated phenol solution, 24 volumes of [chloroform](#) and 1 volume of [isoamyl alcohol](#).

To accurately measure the pH of an equilibrated phenol solution, the following protocol can be used:⁶

For saturated phenols with no chloroform or isoamyl alcohol present:

Mix 2 ml of the organic phase (buffered phenol) with 5 ml [absolute methanol](#) and 13 ml of water. A single phase should result.

For phenol:chloroform:isoamyl alcohol solutions:

Mix 2 ml of the organic phase with 6 ml of [absolute methanol](#) and 10 ml water.

The resulting mixture should be read with a pH meter against a calomel (mercury/mercuric chloride) reference electrode. A silver/silver chloride reference electrode will interfere with the accurate reading of Tris containing solutions. This method has an accuracy of 0.2 pH units.

Availability:

Catalog Number	Description	Size
800672	Phenol, Ultra Pure	100 g
818048		500 g
800673		1 kg
194011	Phenol, molecular biology reagent	25 g
		100 g
		500 g
191423	Phenol, ACS Reagent Grade	500 g
		1 kg

Also Available:

Catalog Number	Description	Size
802528 802529	Phenol:Chloroform Saturated Solution, pH 6.2	200 ml 400 ml 500 g
193814	Chloroform, ACS Reagent Grade	100 ml 500 ml 1 liter
194059	Isoamyl alcohol, ACS Reagent Grade	100 ml 500 ml
819620 819623 819638	Tris, Ultra Pure	500 g 1 kg 5 kg
816116 816124 816100	Tris Hydrochloride	100 g 500 g 1 kg
806443 806444 806445	2-Mercaptoethanol	25 g 100 g 500 g
155388	Methanol, absolute	250 ml

References:

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- *Can. J. Chem.*, v. **37**, 1294 (1959).
- Gottesfeld, J.M., et al., "The inhibition of deoxyribonuclease I by hydroxybiphenyls." *Biochim. Biophys. Acta*, v. **228(2)**, 365-386 (1971).
- Kleinhenz, E.A. and Cohen, S.B., "Accurate determination of pH in organic phenol and phenol:chloroform." *Biotechniques*, v. **10(6)**, 740-741 (1991).