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ProductInformation

Phenol
Sigma Ultra
Product Number **P 5566**
Storage Temperature 2-8 °C

Product Description

Molecular Formula: C_6H_6O
Molecular Weight: 94.11
CAS Number: 108-95-2
Melting Point: 40.85 °C[1]
Boiling Point: 182 °C[1]
Extinction coefficient: E^{mM} (ethanol) = 1.45 (276 nm), 1.91 (271 nm), 6.03 (218 nm)[2]
pK_a: 10.0 (25 °C)[1]
Synonyms: Carbolic acid, Hydroxybenzene, Benzenol

Trace elemental analyses have been performed on the SigmaUltra Phenol. The Certificate of Analysis provides lot-specific results. SigmaUltra Phenol is for applications which require tight control of elemental content.

Phenol may be used as a preservative and as a reagent in chemical analysis. It is used in the manufacture of colorless or light-colored artificial resins, many medical and industrial organic compounds and dyes.[1]

Phenol is used in classic DNA extractions for removing protein. The best phenol for nucleic acid extractions is freshly distilled such as Product No. P 1037. If phenol becomes discolored, it is very likely oxidized and unsuitable for nucleic acid work. Oxidation products make DNA resistant to hydrolysis by DNase I. One such oxidation product is o-hydroxybiphenyl. Inhibition arises from direct action on the DNA rather than on the enzyme, due to hydrogen bonding between bases of the nucleic acid. Several hydroxy biphenyls, including this one, have been studied.[3]

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.

To prepare a phenol:choloroform:isoamyl alcohol reagent for DNA isolation, the phenol should initially be melted in a 45-50 °C water bath, and then the other reagents can be added.[4] Hydroxyquinoline is typically added to this mixture as an antioxidant, but can be omitted, if necessary. However, the solution will last longer if it is added. In addition, the liquified phenol can be saturated with buffers to adjust the pH to the required value.

To accurately measure the pH of a buffered phenol solution, the following procedure should be followed:[5] For saturated phenol solutions (no chloroform: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 of water. The resulting mixture should be read with a pH meter against a calomel (mercury/mercuric chloride) reference eletrode. A silver/silver chloride reference electrode will interfere with the accurate reading of Tris-containing solutions. Use of pH strips will not give accurate measurements. This method has an accuracy of 0.2 pH units.

Precautions and Disclaimer
For Laboratory Use Only. Not for drug, household or other uses.

Preparation Instructions
This product is soluble in water (50 mg/ml).

References

1. The Merck Index, 12th ed., Entry# 7390.
2. Can. J. Chem., **37**, 1294 (1959).
3. Gottesfeld, J. M., et al., The inhibition of deoxyribonuclease I by hydroxybiphenyls. Biochim. Biophys. Acta, **228(2)**, 365-386 (1971).
4. Molecular Cloning: A Laboratory Manual, 3rd ed., Sambrook, J. F., et al., Cold Spring Harbor Laboratory Press (Cold Spring Harbor, NY: 2001), B4 (Appendix).
5. Kleinhenz, E. A., and Cohen, S. B., Accurate determination of pH in organic phenol and phenol:chloroform. Biotechniques, **10(6)**, 740-741 (1991).

HLD 8/03