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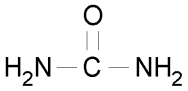
ProductInformation

Urea Solution
8 M after reconstitution

Catalog Number **U4883**
Store at Room Temperature

CAS RN 57-13-6
Synonyms: Carbamide, Carbonyl diamide



Product Description

Molecular Formula: CH_4N_2O
Molecular Weight: 60.06

After addition of 16 ml of water, each bottle will contain 25 ml of an 8 M Urea Solution. This product has been tested and found suitable for use in electrophoresis.

Urea is a chaotropic agent useful for the solubilization and denaturation of proteins.[1-3] Urea is also useful for renaturation of protein samples previously denatured with 6 M guanidine hydrochloride (inclusion bodies[4,5]).

Urea is commonly used for sample preparation prior to electrophoretic methods such as isoelectric focusing (IEF), SDS polyacrylamide gel electrophoresis (SDS-PAGE) and two-dimensional electrophoresis (2DE).[2,3,6-8] Preparing samples for 2DE typically involves solubilization, denaturation, and reduction in order to completely disrupt the interactions between the proteins.[2] Urea is also used to facilitate enzymatic digestion of proteins for analysis by mass spectrometry.

In the presence of heat, urea will break down to form isocyanate, which can lead to carbamylation of the proteins. Isocyanates react with the amino terminus of proteins, preventing N-terminal sequencing. Isocyanates also react with the side chains of lysine residues resulting in a protein that is unsuitable for many enzymatic digests. In addition, carbamylation often leads to confusing results from peptides having unexpected retention times and masses.[9] Care must be taken with 2DE protein samples containing urea to avoid heating the sample above 30 °C. Protein modification due to carbamylation will result in artifacts on a 2DE gel.

When performing enzymatic protein digests it is important to dilute or remove urea first. Even though some enzymes will tolerate small amounts of urea, the elevated temperature used for most reactions may lead to carbamylation during the course of the digest. The urea can be removed prior to digestion by fast reverse phase chromatography, spin columns, or dialysis.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions:

8 M Urea Solution - add 16 ml of deionized water or the buffer of choice to the contents of the bottle. The final volume should be 25 ml.
Note: The solution will initially become cold to the touch. Warm the bottle at 20–25 °C for ~30 minutes, while mixing periodically to ensure complete dissolution. A 30 °C water bath will aid in the dissolution of the powdered urea. Do not allow the temperature of the solution to rise above 30 °C, since cyanates may begin to form that will be detrimental to the proteins.

The 8 M Urea Solution may be used to prepare solutions of lower concentrations as appropriate (see Table 1).

Table 1.
Dilution Table – These dilutions are based on a 25 ml starting volume of 8 M Urea Solution.

Desired Molarity	Volume of water to add to 25 ml of prepared 8 M Urea Solution	Final Volume
8 M	0.0 ml	25.0 ml
7 M	3.6 ml	28.6 ml
6 M	8.3 ml	33.3 ml
5 M	15.0 ml	40.0 ml
4 M	25.0 ml	50.0 ml

Storage/Stability
The product should be stored at room temperature.

Urea solutions should always be **freshly prepared and used**, as solutions of urea may develop a significant concentration of reactive cyanate ions upon standing.

Procedure
Protein Denaturation or Solubilization of Cell Paste

1. Add 1 ml of 8 M Urea Solution per 0.1 g of wet cell paste.
2. Vortex the suspension for 2 minutes.
   Note: Sonicating the sample may increase solubilization.
3. Centrifuge the samples at 15,000 × *g* for 10 minutes at room temperature to remove cell debris.
4. After centrifugation, carefully remove the supernatant. The supernatant contains the denatured, soluble proteins.

References

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