Product Information

79266 Trimethylphenylammonium hydroxide solution

~0.5 M in methanol, for GC derivatization, LiChropur®

Storage temperature: 2-8°C

TMAH (trimethylanilinium hydroxide),* 0.5 M in methanol, is an esterification reagent used to form methyl derivatives, especially of molecules having replaceable protons attached to nitrogen (N-methyl derivatives). It is particularly useful for methylating barbiturates, sedatives, xanthine bases, phenolic alkaloids, dilantin, anticonvulsants, and fatty acids. TMAH is a preferred reagent for derivatizing barbiturates, because most are nitrogen-bearing molecules. TMAH can be used in flash alkylation, in which the analyte is derivatized in the injection port of the gas chromatograph, typically at an injection port temperature of 200-300°C. Barbiturates, biological fluids, and thermally stable fatty acids are suited to this type of derivatization. Flash methylation with TMAH eliminates degradation of barbiturates and their N-methyl derivatives, especially in the derivatization of phenobarbital.

**Note*: The acronym TMAH is used for trimethylanilinium hydroxide, but TMAH also is used for tetramethylammonium hydroxide, $(CH_3)_4NOH$. Always verify the reagent by the full name.

Features/Benefits

- Provides convenient, fast, quantitative derivatization of nitrogen-bearing molecules.
- Preferred reagent for derivatizing barbiturates (except meprobamate, which is analyzed as a free base).

Typical Procedure

This procedure is intended to be a guideline and may be adapted as necessary to meet the needs of a specific application. Always take proper safety precautions when using a esterification reagent.

Prepare a reagent blank (all components, solvents *except sample*), following the same procedure as used for the sample.

1. Extract sample with appropriate solvent, or weigh 1-10 mg of sample into a reaction vessel. If appropriate dilute neat sample with solvent.
2. Add TMAH reagent (begin with equal amounts of sample and TMAH; up to 1000-fold molar excess of reagent has been used).
3. Analyze a 1 µL aliquot of the material by GC (direct injection).

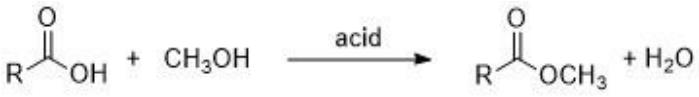
   Flash alkylation
   Draw 1 µL TMAH, then 1µL sample, then 1 µL TMAH into a 10 µL syringe. Inject into the heated GC injection port. Alternatively, either the first or the second aliquot of TMAH can be omitted.

Derivatization times vary widely, depending upon the specific compound(s) being derivatized. If derivatization is not complete (chromatogram peak is smaller than expected), add additional reagent or reevaluate the injection port temperature. If unexpected peaks are present in the chromatogram, reevaluate the amount of TMAH added (excess reagent could cause sample decomposition), the time allowed for the sample and reagent to mix, and the injection port temperature. A low injection port temperature can produce poor responses (incomplete reaction) and/or peak tailing. A high injection port temperature can break down analytes, or produce ghost peaks, poor peak symmetry, baseline drift, or poor response.

Note: Use of TMAH can cause ring opening in ring-containing pharmaceuticals. The extent of this occurrence is affected by the concentration of the reagent and the length of the reaction time. A buffering method has been reported to minimize this problem.[1]

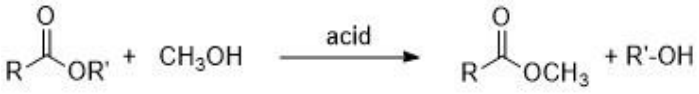
Mechanism[2-4]

Esterification



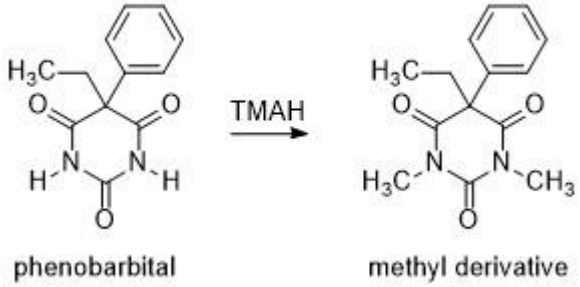
Esterification involves heating the molecule bearing the amino, hydroxyl, carboxyl, or other reactive group with an acid catalyst in an alcohol solvent. The catalyst protonates an oxygen atom or the nitrogen atom of the reactive group, making the molecule much more reactive to nucleophiles. An alcohol molecule then combines with the protonated group, to yield the ester product (*e.g.* R-COO-CH_3 , R-N-CH_3) with loss of water. Esterification is a reversible reaction. Water must be removed to drive the reaction to the right and obtain a high ester yield. A chemical reagent can be used to remove water as it is formed or, if the reaction is conducted at a temperature above 100°C, water may distill off as it is formed. 2,2-dimethoxypropane can be introduced into the reaction mixture to react with the water, yielding acetone. Other water scavengers are anhydrous sulfuric acid and graphite bisulfate. In transesterification, the alcohol is displaced from the ester by another alcohol (*e.g.* methanol) in a process similar to hydrolysis (the second alcohol is used instead of water), forming a new ester.

Transesterification



Transesterification also is an equilibrium reaction. To shift the reaction to the right, it is necessary to use a large excess of the second alcohol, or to remove one of the products from the reaction mixture. Conversion is maximized if excess alcohol is used. The conversion rate also is influenced by the reaction temperature – the reaction generally is conducted near the boiling point of the alcohol.

Action of TMAH



phenobarbital methyl derivative

Storage/Stability

Recommended storage conditions for the unopened product are stated on the label. Store in a bottle or ampule in a cooler in a dry, well ventilated area. Use only in a well ventilated area. Keep away from ignition sources. **Before reuse, validate that your storage conditions adequately protected the reagent.**

References

1. Skinner et al., *Anal. Chem.* **1973**, *45*, 574; Osiewicz et al., *J. Chromatogr.* **1974**, *88*, 157.
2. K. Blau and J. Halket, *Handbook of Derivatives for Chromatography* (2nd ed.), John Wiley & Sons, New York, 1993.
3. D.R. Knapp, *Handbook of Analytical Derivatization Reactions,* John Wiley & Sons, New York, 1979.
4. *Bailey's Industrial Oil and Fat Products* (5th ed.) Vol. 5, John Wiley & Sons, New York, 1995.

Additional Reading

„Comprehensive Drug Screening In Blood for Detecting Abused Drugs or Drugs Potentially Hazardous for Traffic Safety", P. Lillsunde, L. Michelson, T. Forsstrom, T. Korte, E. Schultz, K. Ariniemi, M. Portman, M.L. Sihvonen, T. Seppala, *Forensic Sci. Int.* ***1996***, *77* (3), 191-210; *„Intra-Injector Formation of Methyl Esters from Phenoxy Acid Pesticides*", I. Brondz, I. Olsen, *J. Chromatogr.* **1992**, *598*, 309-312.

Precautions and Disclaimer

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