anatrace

Technical Bulletin 130

Anapoe Detergents

Most polyoxyethylene detergents used in the early studies of membrane proteins were developed for large industrial applications. Today an increasing number of well defined, highly purified detergents, designed specifically for the extraction of membrane proteins in their active state, are available. Detergents such as Dodecyl−β−D-Maltopyranoside and Octyl−β−D-Glucopyranoside have replaced many polyoxyethylene detergents. Yet for some proteins, the polyoxyethylene detergents remain very useful—particularly for routine extraction procedures.

Polyoxyethylene detergents are available in a variety of structures and under an often confusing list of trade names, such as Triton®, Tween®, Genapol®, Brij®, Thesit®, Lubrol®, etc. In addition to the confusion caused by trade names, industrial detergents are often a mixture of closely related detergents that may vary from lot to lot. They may also contain additives and contaminants which result in undesirable effects during protein extraction. One such contaminant are peroxides, which can increase in concentration upon aging of the detergent solution[1].

The "aging" of polyoxyethylene detergents results from the tendency of ethers to react with oxygen to form peroxides. Light accelerates this process. In samples of polyoxyethylene detergents, hydrogen peroxide and organic peroxides with a variety of structures may be found [2-4]. In fact, the concentration of hydrogen peroxide can be as high as 0.2%[5]. Conversely, peroxides may react with detergent molecules in solution, resulting in the presence of several undesirable derivatives[2].

The presence of peroxides during extraction of membrane proteins can result in inactivation and/or degradation of biological materials[4]. Sulfhydryl groups are readily oxidized by peroxides and such oxidation induces protein aggregation and inactivation[6-8].

Peroxides can also interfere in biochemical assays[2, 9-10]. Peroxides are likely responsible for the high blanks noted by Heath and Tappel when measuring lipid peroxides in detergent-solubilized membrane components[9]. Even protein determinations can be affected, as noted by Stutzenberger for the Coomassie blue dye-binding assay, bicinchoninic acid method, and the Folin phenol method[10].

To Make Your Use of Polyoxyethylene Detergents as Trouble-Free as Possible, Anatrace Offers Them in Purified Form

These detergents have been chromatographically purified to contain less than 20 µM of equivalent peroxide in a 10% solution. This specification is often an order of magnitude lower than listed by other manufacturers.

Anapoe® detergents from Anatrace® are prepared from industrial detergents which are often mixtures of closely related detergents. Even though Anapoe detergents are chromatographically purified, they are not necessarily homogenous. Homogenous polyoxyethylene detergents available through Anatrace are:

Product No.	Detergent
T350	Tetraethylene Glycol Monooctyl Ether (C_8E_4), Anagrade®
P350	Pentaethylene Glycol Monooctyl Ether (C_8E_5), Anagrade
P340	Pentaethylene Glycol Monodecyl Ether ($C_{10}E_5$), Anagrade

Anapoe Detergents:

Common Polyoxyethylene Detergents	Anapoe Version	Product No.
Tween 20	Anapoe-20	APT020
Brij-35	Anapoe-35	APB035
Brij-58	Anapoe-58	APB058
Tween 80	Anapoe-80	APT080
$C_{10}E_6$	Anapoe-$C_{10}E_6$	APO106
$C_{10}E_9$	Anapoe-$C_{10}E_9$	APO109
$C_{12}E_8$	Anapoe-$C_{12}E_8$	APO128
$C_{12}E_9$	ANAPOE-$C_{12}E_9$	APO129
$C_{12}E_{10}$	Anapoe-$C_{12}E_{10}$	AP1210
$C_{13}E_8$	Anapoe-$C_{13}E_8$	APO138
Nonidet P40 Substitute	Anapoe-NID-P40	APND40
Triton X-100	Anapoe-X-100	APX100
Triton X-114	Anapoe-X-114	APX114
Triton X-305	Anapoe-X-305	APX305
Triton X-405	Anapoe-X-405	APX405

Structure:

$CH_3(CH_2)yO(CH_2CH_2O)xH$

y=7, x=4, tetraethylene glycol monooctyl ether
y=7, x=5, pentaethylene glycol monooctyl ether
y=7, x=6, hexaethylene glycol monooctyl ether
y=9, x=5, pentaethylene glycol monodecyl ether
y=9, x=6, pentaethylene glycol monodecyl ether
y=9, x=9, nonaethylene glycol monodecyl ether
y=11, x=8, octaethylene glycol monododecyl ether
y=11, x=9, nonaethylene glycol monododecyl ether
y=11, x=10, decaethylene glycol monododecyl ether
y=12, x=8, octaethylene glycol monotridecyl ether

Supplied in:

Each polyethylene detergent is supplied in a 10% aqueous solution under argon gas in the following sizes:

50 ampules containing 1 ml of a 10% solution
5 ampules containing 10 ml of a 10% solution
10 ampules containing 10 ml of a 10% solution
500 ml of 10% solution in a screw cap bottle

Storage:

The ampules should be refrigerated at 0-4°C when received. Once opened, transfer the unused portion to a glass tube or bottle and seal under inert gas to prevent the formation of organic peroxides. The sealed tube should then be refrigerated at 0-4°C.

Sample Preparation:

Detergents are shipped as weight fraction aqueous solutions. Generally, one gram of detergent is dissolved in one gram deionized water for a 50%(w/w) solution. Precision of the solution is 1%. The density for the aqueous detergent solution is 0.996 + 0.02 grams/ml at 20-25°C.

Recovery of Excess Detergent:

Excess detergent can be recovered by several means. Extraction, rotovapping, and lyophilization are a few common tools used for this purpose. It should be noted that impurities may be recovered along with the detergent. Organic peroxides may also form during the recovery process.

Analysis:

Detergent concentration and purity can be determined by HPLC analysis. Peroxide content can be determined using the Pierce PeroXOquant Quantitative Peroxide Assay (Catalog # 23280).

References:

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