

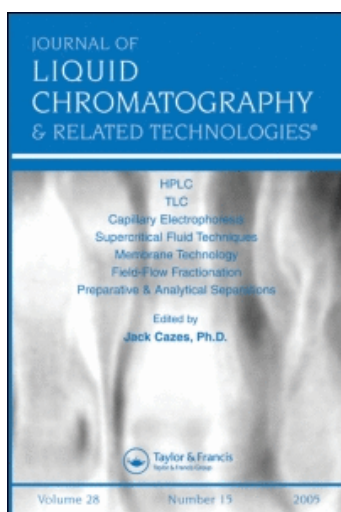
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### Degradation of High Purity Water on Storage

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DEGRADATION OF HIGH PURITY WATER ON STORAGE

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ABSTRACT

Experimental results are presented which indicate that the quality of high purity water degrades with storage time. HPLC trace enrichment techniques were used to monitor the amount of organics in water stored in various types of containers. Starting with initial, high purity water, it was shown that organics could be detected in as little as one hour after storage in plastic containers. Organics could also be detected if the water was stored in glass.

INTRODUCTION

The purpose of this note is to authenticate the fact that the quality of high purity water degrades with storage; that this degradation can be detected within hours; that the degradation generally increases with time, and that the storage container can influence the rate of degradation. This work illustrates the little realized fact that high quality water is a very powerful solvent, and that high quality water can leach materials of construction out of storage containers.

As techniques for analysis in general become more sophisticated and sensitive, the quality of the reagents used for diluting, for controls, for blanks, for standards, etc. becomes increasingly more critical, and in some cases may be the limiting factor in an analytical procedure (1). Impurities found in reagents are particularly important in those analytical procedures where trace concentrations are being measured.

Water has always been the most ubiquitous reagent in many types of analyses, and this is especially so in measurements performed on biological systems. Water quality is also a dominating factor in biological work because water is a universal solvent for biomolecules and cells where impurities can affect enzymatic activities, bacterial, mammalian and plant cell growth. The fact that water quality is important in general analytical and biochemical measurements is noted by the water quality standards that have been established by a number of organizations (ACS, ASTM, NCCLS, CAP). Systems are commercially available which can produce high purity water; however, storing this water for future use results in a degradation in the original quality that has not been readily documented.

#### EXPERIMENTAL

The design of our work was to simulate common laboratory practices that are routinely used to store water, and to monitor the general organic quality of this water as a function of time upon storage. No arduous or extraordinary measures were taken to preserve the quality of the initially pure water other than those which would normally be practiced in a typical biochemical or biological laboratory.

Water quality was measured qualitatively via HPLC using a trace enrichment technique (2). In this procedure, 40ml of test water is loaded into a reverse phase C18 column (Waters Associates) at 2ml/min. A linear gradient of 100% water to 100% acetonitrile over 20 minutes is then run to elute organics adsorbed to the column. The initial high purity water used was produced from a Milli-Q<sup>R</sup> system with an Organex-Q<sup>R</sup> cartridge (Millipore Corporation). Water from this system has been shown to be low in organics content and suitable for LC work(1).

The following materials were tested for storage containers: a 4 liter polyethylene bottle (Scientific Products, B-75-10-128); a 4 liter polypropylene bottle (B-75-13-128), and a 4 liter brown glass bottle (J. T. Baker) that is normally used for shipping LC grade water. In order to prevent inadvertent contamination due to handling, a Teflon<sup>R</sup> tube was inserted through the closed tops of the respective containers so the water could be introduced to the LC columns with little or

no extra handling or manipulation. Each container was initially rinsed thoroughly with water followed by rinsings with the Milli-Q<sup>R</sup> water. Milli-Q<sup>R</sup> with the Organex-Q<sup>R</sup> cartridge water was also used as the mobile phase in all LC runs. Acetonitrile was LC grade (Burdick and Jackson). Detection of organics eluted by the gradient was performed using a UV detector at both 254nm and also 214nm (Waters Associates). Water was stored and measurements were taken at various time intervals for four weeks.

After the initial storage test was completed, the test water was removed, the container recleaned, and a new test cycle was restarted lasting one week. The purpose of this second trial was to determine if the first load of high purity water and storage for four weeks had exhausted all of the leachable material from the containers, or whether more material could be leached by a fresh batch of water. It is common in biochemical laboratories to "condition" storage containers by exhaustingly rinsing any leachates out of plastic containers for several weeks.

#### RESULTS

The results of the storage experiments as a function of time for polyethylene, polypropylene and glass containers are shown in Figures 1, 2, and 3 respectively. Chart speed was 0.60cm/minute and AUFS is 0.02. The lower most LC trace in each set is for the Milli-Q<sup>R</sup> water that was initially placed in all these containers. A similar pattern was obtained for the Milli-Q<sup>R</sup> water used as the mobile phase each time as a sample was tested. For both the polyethylene and the polypropylene containers, one can notice a degradation in water quality beginning in as short a time as one hour. Each peak represents presumably one organic species that is being leached from the plastic containers. No effort was made to determine the identification of what organics these various peaks represented. The LC traces for one day, one week and two weeks are similar to that of one hour except that certain peaks are intensified, and the general trend is seen that the amount of contaminants detected increases with time. The LC traces for the water stored in the glass containers show the same general character as for the water in the plastic containers. The peaks, however, are not as pronounced, nor do they appear as rapidly, compared to the corresponding plastic storage systems.

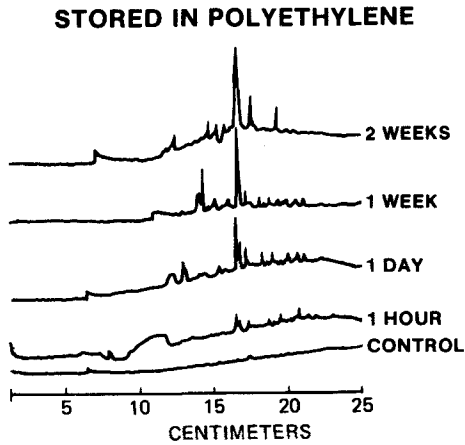


Figure 1: Trace enrichment chromatograms of high purity water stored in a polyethylene container.

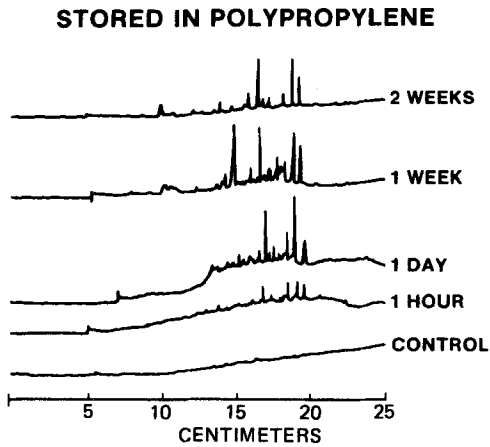


Figure 2: Trace enrichment chromatograms of high purity water stored in a polypropylene container.

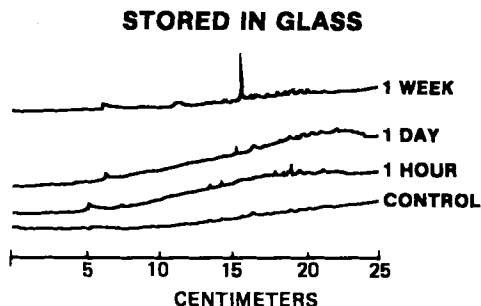


Figure 3: Trace enrichment chromatograms of high purity water stored in a glass container.

LC traces at 214nm showed similar trends with time as those at 254nm. Also, the results of the second trial with fresh, high purity water followed a pattern similar to the first trial.

#### DISCUSSION

Healy (3) showed that doubly distilled water could leach inorganic ions out of Pyrex<sup>R</sup> containers during a storage period of two weeks. Our study has illustrated a corresponding situation for organics in plastic containers. Upon storage, the general quality of initially pure water degrades as measured by the increase in organic constituents.

Although the relative contributions are unknown at this time, there are basically three sources for the organics detected in the LC traces.

1. Plasticizers, monomers, mold release agents and the oligomers used in the construction and manufacture of any container can be leached out by the water.
2. Bacteria can grow in the stored water and excrete waste products, thus contributing to the general organics (4).
3. Organics from other reagents, laboratory personnel's respiration, and shedding from clothing can all diffuse through the air and dissolve in the water.

It is quite possible that a succession of these mechanisms occur as a function of time.

Water quality can play an influential role in analytical LC measurements of biochemical components (5), and also in the growth of tissue culture systems. Even if high purity water is initially produced, that same water quality may not exist a short time later. For those analytical and biological systems that are extremely sensitive to water quality, care should be taken not to store the water, but to use the water as it is produced.

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