

# Suggestions for Cleaning Glassware

## Application Note



Clean glassware is critical for both chemical and biological applications, although the criteria for cleanliness may be different.

### Introduction

Good laboratory technique demands clean glassware, because the most carefully executed piece of work may give an erroneous result if dirty glassware is used. In all instances, glassware must be physically clean; it must be chemically clean; and in many cases, it must be sterile. All glassware must be absolutely grease-free. The safest criterion of cleanliness is uniform wetting of the surface by distilled water. This is especially important in glassware used for measuring the volume of liquids. Grease and other contaminating materials will prevent the glass from becoming uniformly wetted. This in turn will alter the volume of residue adhering to the walls of the glass container and thus affect the volume of liquid measured or delivered. Furthermore, in pipets and burets, the meniscus will be distorted and the correct adjustments cannot be made. The presence of small amounts of impurities may also alter the meniscus.

### Safety Considerations

Eye protection and heavy duty slip-resistant and chemically resistant gloves should be used when washing glassware. Depending on the detergents and cleaning solutions being used an apron and fume hood may also be required. Always check with your Safety Office before using caustic washing solutions.

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## Cleaning PYREX® Glassware

Wash glassware as quickly as possible after use. The longer it is left unwashed the harder it will be to clean. If a thorough cleaning is not possible immediately, disassemble the glassware and put it to soak in water. This is especially important for ground glass stopcocks and stoppers. If glassware is not cleaned immediately, it may become impossible to remove the residue. Do not overload sinks, dishwashers, or soaking bins. Rubber sink and counter mats can help reduce the chance of breakage and resultant injury. Most new glassware is slightly alkaline in reaction. For precision chemical tests, new glassware should be soaked several hours in acid water (a 1% solution of hydrochloric or nitric acid) before washing.

### Glassware Cleaners

When washing, soap, detergent, or cleaning powder (with or without an abrasive) may be used. Cleaners for glassware include Alconox®, Liquinox®, Lux®, Tide® and Fab®. The water should be hot. For glassware that is exceptionally dirty, a cleaning powder with a very mild abrasive action, such as BonAmi®, will give more satisfactory results. The abrasive should not scratch the glass.

During the washing, all parts of the glassware should be thoroughly scrubbed with a brush. This means that a full set of brushes should be available: brushes to fit large and small test tubes, burets, funnels, graduates and various sizes of flasks and bottles. Brushes with wooden or plastic handles are recommended as they will not scratch or abrade the glass surface. Motor driven revolving brushes are valuable when a large number of tubes or bottles are processed. Do not use cleaning brushes that are so worn that the brush spine hits the glass. Serious scratches may result. Scratched glass is more prone to break during experiments. Any mark in the uniform surface of glassware is a potential breaking point, especially when the piece is heated or used in vacuum applications. Do not allow acid to come into contact with a piece of glassware before the detergent (or soap) is thoroughly removed. If this happens, a film of grease may be formed.

### Other Cleaning Methods

**Caution!** The following methods can cause serious damage to the eyes, mucus membranes, skin and lungs and should only be undertaken by people trained in their proper use and fully equipped with heavy duty slip resistant and chemically resistant gloves, eye protection, lab coat, apron and, when appropriate, a fume hood.

If glassware becomes unduly clouded or dirty or contains coagulated organic matter, it must be cleansed with more aggressive and potentially dangerous cleaning solutions often using concentrated acids or bases. Special types of precipitates may require removal with nitric acid, aqua regia or fuming sulfuric acid. These are very corrosive substances and should be used only when required. Chromic acid, commercially available as Chromerge® cleaning solution, dissolved in concentrated sulfuric acid is also a very powerful cleaner. However, the use of Chromerge or other chromate-based cleaning solutions is not recommended in many research laboratories because the chromium ions are highly toxic to the environment and pose a severe waste disposal problem, even in small quantities. In addition, the chromium present in Chromerge is considered to be a potent human carcinogen. A safer alternative is the use of NoChromix® cleaning solution, containing hydrogen peroxide, which is also made up in sulfuric acid. While still very caustic, it does not contain any toxic chromium. Please follow the manufacturer's direction for use of this product.

When NoChromix solution is used the glassware may be rinsed with the cleaning solution or it may be filled with it and allowed to stand. The length of time it is allowed to stand depends on the amount of contamination on the glassware. Relatively clean glassware may require only

a few minutes of exposure; if debris is present, such as blood clots, it may be necessary to let the glassware stand all night. Due to the intense corrosive action of the NoChromix solution, it is good practice to place the stock bottle, as well as the glassware being treated, in flat glass pans or pans made from plastic polymer determined compatible with the concentration of NoChromix you are using. Extra care must be taken to be sure all caustic cleaning solutions are disposed of properly.

### Removing Grease

Grease is best removed by boiling in a weak solution of sodium carbonate. Acetone or any other fat solvent may be used. Strong alkalis should not be used. Silicone grease is most easily removed by soaking the stopcock plug or barrel for 2 hours in warm decahydronaphthalene. Grease can also be removed from ground joints by wiping with a paper towel soaked in acetone or other appropriate solvent. Use a fume hood to minimize exposure to the fumes.

Drain and rinse degreased glassware with acetone or use fuming sulfuric acid for 30 minutes. Be sure to rinse off all of the cleaning agents.

### Rinsing

It is imperative that all soap, detergents and other cleaning fluids be removed from glassware before use. This is especially important with the detergents, slight traces of which will interfere with serologic and cell culture applications.

After cleaning, rinse the glassware with running tap water. When test tubes, graduates, flasks and similar containers are rinsed with tap water, allow the water to run into and over them for a short time, then partly fill each piece with water, thoroughly shake and empty at least six times. Pipets and burets are best rinsed by attaching a piece of rubber tubing to the faucet and then attaching the delivery end of the pipets or burets to a hose, allowing the water to run through them. If the tap water is very hard, it is best to run it through a deionizer or reverse osmosis system before using.

Next rinse the glassware in a large bath of high purity or distilled water. Then do a final individual rinse of each item with high purity water. To conserve water, use a five gallon bottle as a reservoir. Store it on a shelf near your clean-up area. Attach a siphon to it and use it for replenishing the reservoir with used distilled water.

### Cleaning PYREXPLUS® Glassware

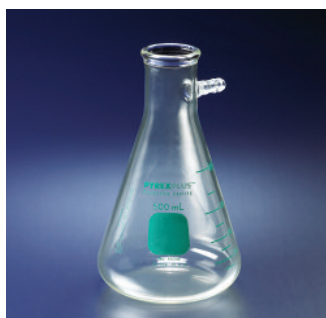
PYREXPLUS glassware is coated with a tough, transparent plastic vinyl layer. The coating, which is applied to the outside of the vessel, helps prevent exterior surfaced abrasion. It also helps minimize the loss of contents and helps contain glass fragments if the glass vessel is broken.

Any non-abrasive glassware detergent may be used for hand or automatic dishwasher cleaning. If using a dishwasher or glassware dryer, care should be taken to be sure the drying temperature does not exceed 110°C (230°F). Exposure to dry heat should be minimized.

Avoid brushes and cleaning pads which could abrade the glass or damage the coating. If using a chromic acid cleaning solution minimize contact of the solution with the coating.

PYREXPLUS glassware can be repeatedly autoclaved using liquid or dry cycle sterilization which involves no vacuum or low vacuum (less than 5 inches mercury).

Sterilization time should not exceed 15 minutes at 121°C (250°F). Drying time should not exceed 15 minutes at 110°C (230°F). The actual cavity temperature of the autoclave should be checked to be sure the autoclave temperature does not exceed the recommended sterilization and drying temperature. Vessels should not be allowed to touch each other during autoclaving.



PYREXPLUS filtering flasks provide an extra layer of safety when used for vacuum aspiration.

It is better to use a slow cool cycle for venting rather than fast venting to reduce the chance that air pockets will form between the glass and the coating. Should the coating appear clouded due to dissolved moisture, simply let dry overnight at room temperature or briefly heat to 110°C (230°F).

### Cleaning PYREX® Fritted Ware

A new fritted filter should be washed by suction with hot hydrochloric acid and then rinsed with water before it is used. This treatment will remove loose particles of foreign matter such as dust. It is advisable to clean all PYREX fritted filters as soon as possible after use. This will prolong their life.

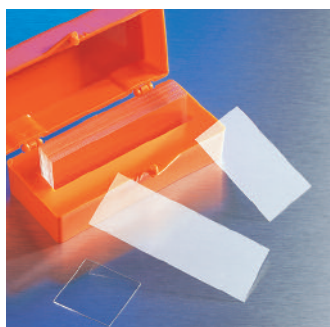
Many precipitates can be removed from the filter surface simply by rinsing from the reverse side with water under pressure not exceeding 15 lb/sq. in. Drawing water through the filter from the reverse side with a vacuum pump is also effective. Some precipitates tend to clog the pores of a fritted filter and may require special cleaning solutions (see Table 1).



Fritted PYREX glassware requires special cleaning procedures to ensure the filter frits do not clog.

**Table 1. Recommended cleaning solutions for PYREX fritted glassware**

Material	Cleaning Solution
Fatty materials	Carbon tetrachloride
Organic matter	Hot concentrated cleaning solution, or hot concentrated sulfuric acid plus a few drops of sodium or potassium nitrite.
Albumen	Hot ammonia or hot hydrochloric acid.
Glucose	Hot mixed acid; H <sub>2</sub> SO <sub>4</sub> + HNO <sub>3</sub> .
Copper, or Iron Oxides	Hot hydrochloric acid plus potassium chlorate.
Mercury Residue	Hot nitric acid.
Silver Chloride	Ammonia or sodium hyposulfite.
Viscose	5 to 10% NaOH, followed by cleaning solution.
Aluminous and Siliceous Residues	2% hydrofluoric acid followed by concentrated sulfuric acid; rinse immediately with distilled water followed by a few milliliters of acetone. Repeat rinsing until all traces of acid are removed.



Corning optically clear cover glasses are available in a variety of sizes and shapes.

### Cleaning Corning® Slides and Cover Glass

It is especially important that microscope slides and cover glass used for the preparation of blood films, bacteriologic smears or cell culture be perfectly clean and free from scratches. Slides should be washed, placed in glacial acetic acid for 10 minutes, rinsed with distilled water and wiped dry with clean paper towels or cloth.

Before use, wash with alcohol and wipe dry. Or the slides, after acid treatment and rinsing, may be placed in a wide jar and covered with alcohol. As needed, remove from the jar and wipe dry.

### Cleaning PYREX Burets

Remove the stopcock or rubber tip and wash the buret with detergent and water. Rinse with tap water until all the dirt is removed. Then rinse with distilled water and dry.

Wash the stopcock or rubber tip separately. Before a glass stopcock is placed in the buret, lubricate the joint with stopcock lubricant. Use only a small amount of lubricant. Burets should always be covered when not in use.



PYREX® Burets caption needed

## Cleaning PYREX® Cell Culture Glassware

Cell culture glassware should first be soaked, then washed and repeatedly rinsed with both tap water and high-quality water, i.e., purified by distillation, deionization or reverse osmosis. Special attention should be given to the source of water used during the washing process. Copper tubing is frequently a source of toxic metallic ions in cell culture systems. To eliminate this problem, appropriate plastic or stainless steel tubing can be substituted. Another source of toxic metallic ions may be the hot water heating system used in glassware washing. A separate glass lined hot water system, located in proximity to the glass washing area, may eliminate this.

A source of toxic heavy metal ions often overlooked are media storage bottles that have been previously used to hold staining solutions for electron microscopy that contain osmium tetrochloride, uranyl acetate or lead nitrate. These metal ions can bind tightly to the glass and may not be released during cleaning unless concentrated acid cleaners are used. When media are then stored in the bottles the metal ions will slowly release from the glass surface back into the medium resulting in cell toxicity. Disposable bottles are recommended for storing these solutions.

Usually only borosilicate (such as PYREX) glassware is recycled. Soft soda lime or flint glasses should be used once and then discarded; repeated use of soft glass may cause leaching of toxic materials into solutions or cultures. It is essential that glassware be thoroughly cleaned and rinsed. Some cleaning agents, such as 7x® cleaning solution, have been specifically developed for washing cell culture glassware and are designed to not leave any toxic detergent residues after rinsing. If a central glass washing service cannot provide the attention necessary to process cell culture grade glassware, then glassware must be washed within individual laboratories. The use of disposable plasticware can eliminate or greatly reduce this problem.

## Cleaning PYREX Culture Tubes

Culture tubes which have been used previously must be sterilized before cleaning. The best general method for sterilization cultures is by autoclaving for 30 minutes at 121°C (15 lb. pressure). Media which solidify on cooling should be poured out while the tubes are hot. After the tubes are emptied, brush with detergent and water, rinse thoroughly with tap water, rinse with distilled water, place in a basket and dry.

If tubes are to be filled with a medium which is sterilized by autoclaving, do not cap tubes until the medium is added. Both medium and tubes are thus sterilized with one autoclaving. Make sure the caps can be autoclaved. Caps with paper liners should not be autoclaved.

If the tubes are to be filled with a sterile medium, plug and sterilize the tubes in the autoclave or dry air sterilizer before adding the medium. Corning also offers a variety of disposable glass and plastic culture tubes that eliminate the need for cleaning.

## Cleaning Pipets

Place pipets, tips down, in a cylinder or tall jar of water immediately after use. Do not drop them into the jar, since this may break or chip the tips and render the pipets useless for accurate measurements. A pad of cotton or glass wool at the bottom of the jar will help to prevent breaking of the tips. Be certain that the water level is high enough to immerse the greater portion or all of each pipet. At a convenient time, the pipets may then be drained and placed in a cylinder or jar of dissolved detergent or, if exceptionally dirty, in a jar of chromic acid cleaning solution. After soaking for several hours, or overnight, drain the pipets and run tap water over and through them until all traces of dirt are removed. Soak the pipets in distilled water for at least 1 hour. Remove from the distilled water, rinse, dry the outside with a cloth, shake the water out and dry.

In laboratories where a large number of pipets are used daily, it is convenient to use an automatic pipet washer. Some of these, made of metal, are quite elaborate and can be connected directly by permanent fixtures to the hot and cold water supplies. Others, such as those made



Corning offers a full selection of both reusable and disposable PYREX glass and plastic storage bottles designed to meet the stringent requirements of cell culture.



Corning offers both reusable and disposable glass culture tubes as well as sterile disposable plastic culture tubes



Corning offers sterile disposable glass and plastic pipets to eliminate the need for washing and sterilization.

with polyethylene, are less elaborate and can be attached to the water supplies by a rubber hose. Polyethylene baskets and jars may be used for soaking and rinsing pipets in chromic acid cleaning solution. Electrically heated metallic pipet driers are also available.

After drying, place chemical pipets in a dust-free drawer. Wrap serologic and bacteriologic pipets in paper or place in pipet cans and sterilize in the dry air sterilizer at 180°C for 2 hours. Pipets used for transferring infectious material should have a plug of cotton placed in the top end of the pipet before sterilizing. This plug of cotton will prevent the material being measured from being drawn accidentally into the pipetting device. Corning also offers a full line of sterile, ready to use disposable glass and plastic pipets to eliminate the need for cleaning and sterilizing.

### Cleaning PYREX Blood Chemistry Pipets

After use, rinse thoroughly with cool tap water, distilled water, alcohol or acetone, and then either.

Dry by suction. (Do not blow into the pipets as this will cause moisture to condense on the inside of the pipet).

To remove particles of coagulated blood or dirt, a cleaning solution should be used. One type of solution will suffice in one case, whereas a stronger solution may be required in another. It is best to fill the pipet with the cleaning solution and allow it to stand overnight. Sodium hypochlorite (laundry bleach) or a detergent may be used. Hydrogen peroxide is also useful. In difficult cases, use concentrated nitric acid. Some particles may require loosening with a horse hair or piece of fine wire. Take care not to scratch the inside of the pipet.



PYREX® Ostwald-Folin blood chemistry pipets must be carefully cleaned to ensure their accuracy.

### Siliconizing Glassware

Siliconizing glass to make its surface very hydrophobic is often used to prevent cells and other biological materials from adsorbing to the glass surface. Siliconizing (also called silanizing) glassware requires coating the clean and dry glass surface with a highly reactive organopolysiloxane which reacts with the glass giving off toxic gases. Treatment should always be done in a fume hood wearing appropriate safety equipment following manufacturers' instructions. Siliconizing reagents for glassware can be obtained from Sigma (Sigmacote® Cat. No. SL-2) or Fisher Scientific (AquaSil™ Siliconizing Fluid).

### Rinsing, Drying and Storing Glassware

Be careful when rinsing or washing pipets, cylinders or burets not to let tips hit the sink or the water tap. Most breakage occurs in this way. Dry test tubes, culture tubes, flasks and other glassware by hanging them on wooden pegs or placing them in baskets with their mouths downward and allowing them to air dry. Alternatively, place them in backsets and dry in an oven. The temperature for drying should not exceed 140°C. (Never apply heat directly to empty glassware used for volumetric measurements. Such glassware should be dried at temperatures of no more than 80°C to 90°C.) Before placing glassware in a basket, cover the bottom of the basket with a clean folded towel or clean piece of cloth. This prevents the mouths of the tubes from becoming dirty.

Dry burets, pipets and cylinders by standing them on a folded towel. Protect clean glassware from dust. This is done best by plugging with cotton, corking, taping a heavy piece of paper over the mouth or placing the glassware in a dust-free cabinet.

When storing, place pieces in racks designed especially for them. Be sure pieces do not touch each other, to avoid inadvertent mechanical damage. Do not store glassware close to the front edge of shelves.

Do not store alkaline liquids in volumetric flasks or burets. Stoppers or stopcocks may stick.