

PolySeed®

Technical Report

Carboys & Contamination

There are three test controls in the BOD5 test: dilution water *blanks* that check the quality of unseeded dilution water and cleanliness of the BOD bottles; seed *controls* that determine how much seed should be added to samples; and *GGA standard checks* that check seed effectiveness.

Although the controls have several testing variables in common, only one of these common variables has the power to negatively affect all three sets of controls: the *Dilution Water*. Other shared test components, such as glassware and user technique, can impact various aspects of the test in isolation. Dilution water, on the other hand, never has an isolated impact. Contaminated dilution water will render the *entire* test *Invalid, without exception*.

The good news is that contaminated dilution water is an easy problem to solve! Often, the root problem isn't bad source water....it isn't old reverse osmosis cartridges in need of replacing....and no, we can't blame it on the guy who keeps sneezing in our general vicinity. The most common cause of contaminated dilution water is actually very simple: a *ditty carboy*.

Having "clean" dilution water is very important when running the BOD5 test and clean dilution water starts with a clean carboy. A properly maintained carboy plays a big role in achieving good results. Read below for a few tips on how to keep your carboy clean, prevent contamination and keep the all-encompassing dilution water variable in check.

Please Note: Dilution Water is sometimes referred to as "BOD Water" or "Nutrient Water"*

Dilution Water Tips: How to prevent contamination

Use separate tubing/air diffusers to aerate dilution water

- The BOD₅ test requires two sets of aeration tubing: one set dedicated to aerating the dilution water, and the other dedicated to aerating the seed solution.
- Store tubing in separate bags; clearly label storage bags "seed solution" and "dilution water" to avoid mix-ups.
- Replace both sets of tubing and diffusers often to avoid contamination and mold growth.

Keep a separate carboy just for storing dilution water

- Designate one carboy for “dilution water only”
- Clearly label the carboy to avoid mix-ups
- Never mix seed or chemical stock solution in the dilution water carboy

Don't let the neck of the BOD bottle touch the carboy spigot

- Because the carboy spigot can become contaminated by dirty gloves, it can become the dirtiest part of the carboy.
- It is good practice to keep the neck of the BOD bottles from touching the spigot to prevent cross contamination

Thoroughly clean your spigot at least once a month

- Sanitize the carboy spigot at least once a month.
- To sanitize, soak the spigot in a 10% HCl acid solution for at least one hour.
- To ensure complete sanitization, always disassemble the spigot before soaking.
- Triple rinse the spigot with DI water and allow the spigot to air dry.

The Do's and Don'ts of cleaning a carboy

DO triple rinse the carboy with DI water before and after each use.

DON'T use soap! Soap residue inhibits microbial action. The bacteria will not eat, breathe, or reproduce as much as they should, causing low depletions.

DON'T use bleach! Clorox is an alkaline from chlorine (HOCl). Many labs use bleach as a cleaning agent, but it is *not* recommended for the sanitization of BOD equipment. Even a very small amount of residual chlorine can cause significant fluctuations in the BOD test.