



Frequently Asked Questions for Performa® DTR Cartridges

Sample Recovery

1. **What is the maximum sample volume?** The recommended maximum volume is 20 μl . Larger volumes loaded onto the column may result in dye terminator and salt contamination.
2. **What is the minimum sample volume?** The minimum recommended volume is 10 μl . Smaller volumes loaded onto the column will result in low signal strength, loss of 5' sequence, and shorter read lengths.
3. **What will be the final volume of the purified sample?** The final volume of the purified sample should be approximately 1-4 μl higher than that of the initial sample.
4. **My reaction volumes are <10 μL . Do you recommend bringing the sample volume to 10 μL ?** Yes. If you are running 5 μl reactions, adjust the sample volume to at least 10 μl .
5. **I centrifuged the cartridge at a speed much lower than recommended. Can I still recover my sample?** Spin the cartridge at the correct speed for an additional minute. You might need to dry the sample and resuspend it in 10-20 μL water. Even though this procedure might "save" your sample, it will not be optimal and you may need to re-run the sequencing reaction.

General

1. **There is a crack in the gel matrix of the cartridge. Will it affect the cartridge performance?** The crack is a normal result of the manufacturing process of the cartridge and does not affect its performance. Take care to avoid the crack when loading sample drop-wise to the center of the gel column.
2. **What are the recommended spin conditions?** Pre-spin the cartridge at 750 x g for 2 minutes, transfer the cartridge to a clean tube, load the sample onto the cartridge and spin at 750 x g for 2 minutes. If you are using BigDye® v. 3.1, and experiencing dye blobs, pre-spin the cartridges at 850 x g for 3 minutes, transfer the cartridge to a clean tube, load the sample onto the cartridge and spin at 850 x g for 3 minutes.
3. **Can I use BigDye® v. 3.1 without experiencing blobs?**
 - a. Yes, but we recommend that you dilute the BigDye® to 1 μl or less to insure optimum performance. Be sure to use sequencing reaction diluent, rather than water, to reduce the concentration of BigDye® version 3.1 in your sequencing reaction .
 - b. If you are using BigDye® v. 3.1, and experiencing dye blobs, pre-spin the cartridges at 850 x g for 3 minutes, transfer the cartridge to a clean tube, load the sample onto the cartridge and spin at 850 x g for 3 minutes.

Storage

1. **I realized the cartridges are frozen. Will that be a problem?** Frozen cartridges cannot be used. The matrix ruptures and seals the base of the unit resulting in total sample loss.



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2. **My centrifuge starts timing before the rotor has reached full speed, will this affect the time settings I should use?** The centrifugation time recommendations are based on time the centrifuge starts spinning. As most centrifuges that accommodate micro centrifuge tubes accelerate to full speed within seconds, such delays are likely to have minimal impact on performance. If your centrifuge takes more than 15 seconds to reach full speed, it may be necessary to adjust your spin conditions. However, we have not observed that slow acceleration in a microcentrifuge significantly alters column performance.
3. **Why does the chromatogram show dye blobs?** Unincorporated dye terminators co-migrate with sequencing products during electrophoresis. The most common peaks occur roughly 70 bases into the sequencing run, and in cases of severe contamination, additional peaks may appear in the vicinity of base 120, 220, or higher. If you are experiencing difficulties with unincorporated dye terminators after using our purification products, please refer to our Trouble Shooting Guide for suggestions on how to eliminate these problems.
4. **Can I store cartridges at room temperature for short periods of time?** Yes. A few hours on the bench or overnight storage will not damage performance or reduce shelf life as long as the cartridges are snapped shut and the zip seal foil bag is properly closed. We do not recommend long-term storage at room temperature.
5. **Does the gel matrix contain preservatives?**
No, the gel matrix is dispensed in deionized water.