



Troubleshooting Guide for Performa® DTR Cartridges

PROBLEM	POSSIBLE CAUSE	SUGGESTED SOLUTIONS
Signal intensity low	Less than 10 µl of sequencing reaction loaded	Adjust the sequencing reaction to 10-20 µl before loading on the cartridge
	Centrifuge conditions were not optimal	Recalculate rpm to achieve selected speed for your dye conditions
	Sequencing reaction conditions were not optimal	Adjust sequencing reaction parameters; consult sequencing reagent manufacturer's recommendations
Dye blobs	Too large a volume was loaded	Decrease volume of reaction loaded
	Centrifuge conditions were not optimal	<ul style="list-style-type: none"> Recalculate rpm to achieve selected speed for your dye conditions Be certain that the pre-spin is for 3 min at 850 x g for BigDye® v. 3.1 Be certain that the pre-spin is 2 minutes at 750 x g for BigDye® v. 3.0 and other dye chemistries
	Sample was not loaded properly	Load the sample slowly, drop-wise to the center of the gel column, taking care to avoid the crack
	Signal intensity too high	Apply only a portion of sequencing reaction to the cartridge; high signal strengths exaggerate the effect of small quantities of dye terminator in the purified reaction
	BigDye® v. 3.1 was used in sequencing reaction	Make sure you follow BigDye® v. 3.1 protocol: pre-spin at 850 x g for 3 minutes, load sample, spin sample at 850 x g for 3 minutes
Truncation of 5' sequence	Centrifuge conditions were not optimal	Recalculate rpm to achieve selected speed for your dye conditions
	Dye blobs present	See Dye blobs above
	Less than 10 µl of sequencing reaction loaded	Increase the volume of reaction to 10-20 µl before loading on the cartridge



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	Sequencing reaction conditions were not optimal	Adjust sequencing reaction parameters; consult sequencing reagent manufacturer's recommendations
Recovery volume higher than expected	Centrifuge conditions were not optimal	<ul style="list-style-type: none"> Recalculate rpm to achieve selected speed for your dye conditions Be certain pre-spin is 2 minutes at 750 x g for BigDye® v. 3.0 Be certain pre-spin is 3 minutes at 850 x g for BigDye® v. 3.1
	More than 20 µl of sequencing reaction loaded	Decrease volume of sequencing reaction loaded to 10-20 µl
	Cartridges have been stored incorrectly	<ul style="list-style-type: none"> Store cartridges at 4°C Do not allow cartridges to freeze Unopened cartridges in a sealed bag can be left at room temperature for short periods of time
Recovery volume lower than expected	Centrifuge conditions were not optimal	Recalculate rpm to achieve selected speed for your dye conditions
	Less than 10 µl of sequencing reaction loaded	Increase volume of sequencing reaction to 10-20 µl
	Cartridges were not stored correctly	<ul style="list-style-type: none"> Store cartridges at 4°C Do not allow cartridges to freeze Do not allow cartridges to dehydrate; store unopened cartridges in a sealed bag

For additional troubleshooting assistance, please contact Edge BioSystems at:

Telephone: (800) 326-2685 or (301) 990-2685	Fax: (301) 990-0881
Email: info@edgebio.com	Web: http://www.edgebio.com