

ClearBand 6x DNA Loading Dye Blue

Troubleshooting Guide

5 x 1 mL, 5 x 5 mL

Cat No: DLD-6xB, DLD-6xB5

Use this guide for common sample-loading and migration-tracking problems encountered with ClearBand 6x DNA Loading Dye Blue. The dye is a premixed 6x loading buffer for agarose and non-denaturing polyacrylamide gels and contains Xylene Cyanol FF and Bromophenol Blue as tracking dyes.

Before You Start - Quick Checks

1. Use 1 volume of 6x Loading Dye Blue with 5 volumes of DNA sample.
2. Mix the dye and DNA sample thoroughly but gently before loading.
3. Load the prepared sample directly into the gel well.
4. Remember that Xylene Cyanol FF migrates around the 4,000 bp region and Bromophenol Blue around the 400 bp region in agarose gels.
5. Store at 4-8°C or room temperature; the datasheet states stability for 1 year.
6. Use clean tips and avoid cross-contamination between DNA samples.

Problem	Most likely cause	Recommended action
Sample does not sink or is difficult to load	The loading dye may have been added at the wrong sample-to-dye ratio or not mixed uniformly.	Prepare the sample at the specified 1:5 dye-to-sample ratio and mix evenly before loading. Confirm that the full volume enters the well rather than remaining on the pipette tip or well edge.
Tracking dyes appear unusually faint	Too little loading dye may have been added or the sample and dye may not have been mixed completely.	Recheck the 1 volume dye to 5 volumes DNA sample ratio and mix before loading. Compare with a freshly prepared sample if the stock handling history is uncertain.
Tracking dye is excessively intense	Too much 6x dye may have been added relative to the DNA sample.	Keep the final loading dye at the intended 1x working concentration by using the stated 1:5 ratio.
Unexpected dark region near PCR-sized fragments	Bromophenol Blue migrates around the 400 bp region in agarose gels and can visually overlap with fragments in this size range.	Interpret bands near this region with the tracking-dye position in mind. If the shadow interferes with visualization of fragments near 400 bp, consider the Green formulation, which uses Orange G instead of Bromophenol Blue.
DNA sample appears to migrate inconsistently	The loading mixture may have been prepared inconsistently between samples or wells may have been loaded unevenly.	Use the same dye-to-sample ratio for all samples and prepare each loading mixture consistently. Load carefully into intact wells.
Tracking dyes run at positions different from expected	Tracking-dye migration depends on the electrophoresis system and the datasheet positions are approximate reference positions in agarose gels.	Use the stated ~4,000 bp and ~400 bp positions as approximate migration references, not DNA size standards.
Sample spills or diffuses from the well during loading	The pipette tip may have punctured the well or the sample may have been dispensed too quickly.	Load slowly into the center of an intact well and avoid touching or piercing the gel with the tip.
Results differ between agarose and native PAGE	The product is compatible with both agarose and non-denaturing polyacrylamide gels, but tracking-dye migration is matrix dependent.	Use the dye for loading in either system, but do not assume that the agarose-gel tracking positions will be identical in non-denaturing polyacrylamide gels.
Cross-contamination between lanes	The same pipette tip or contaminated handling surface may have contacted multiple samples.	Use a fresh sterile tip for each sample and keep tube rims, pipettes, and the gel-loading area clean.
Performance changes after prolonged storage	The product may have been stored beyond the stated stability period or under inconsistent conditions.	Store at 4-8°C or room temperature as specified and use within the stated 1-year stability period.

Still having trouble? Confirm the 1:5 dye-to-sample ratio, verify storage history, and prepare a fresh loading mixture. Use the tracking-dye positions only as approximate migration references and inspect the gel wells for physical damage or loading errors.