A modified protocol can improve high-GC amplicon performance on the Tapestri Platform

High-GC amplicons (GC > 63%) are often among the lower-performing amplicons across many DNA panels. A modified Barcode Mix can help improve the performance of these amplicons at the single-cell level. In limited testing across four Tapestri runs using the catalog Myeloid panel and the DNA workflow, the modifications improved high-GC-amplicon performance while reducing relative coverage for lower GC amplicons.

Note: Protocol modifications are considered in development and results may vary. Data below is based on observations from 2 runs with the modified protocol and 2 control runs using the Myeloid panel with the DNA only protocol. Control and modified runs were normalized to 80x reads/cell/amplicon per standard depth requirements. Cells with ≥ 10 reads for a given amplicon are considered genotyped.

HIGH-GC (63–70%) % CELLS GENOTYPED

80→92%

11 amplicons · +12 %

EXTREME HIGH-GC (≥70%) % CELLS GENOTYPED

21→95%

4 amplicons · +74 %

PANEL UNIFORMITY

91→87%

12 amplicons · -4%

Figure 1 · % Cells Genotyped vs. GC content

Per amplicon, fraction of cells genotyped (projected ≥10 reads/cell/amplicon at 80x coverage), binned by total amplicon GC. Whiskers span n=2 runs per condition.

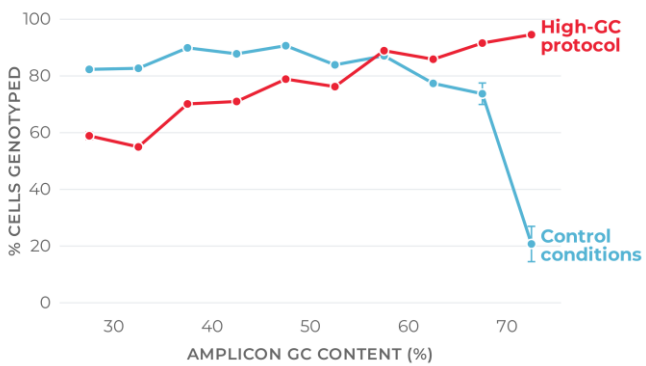
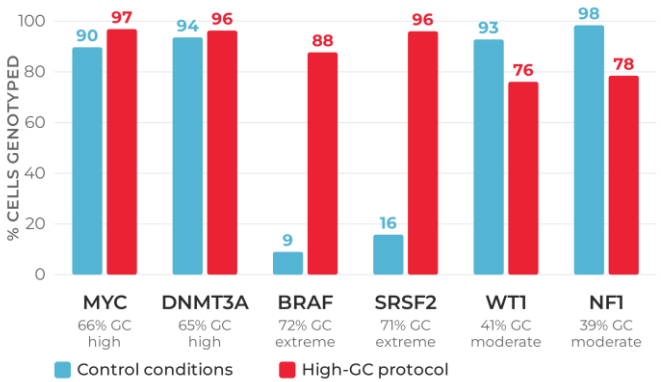


Figure 2 · % Cells Genotyped, by amplicon

Representative high-GC, extreme high-GC, and moderate-GC amplicons.



Key Observation: Extreme high-GC amplicons (BRAF, SRSF2) show a significant increase in performance across most cells with the high-GC protocol modifications. The trade off is a relative decline in coverage for moderate-GC amplicons. Overall observed panel uniformity is reduced as the panel's highest-GC amplicons become its best performing. High-GC protocol conditions may require optimization, which may include primer concentration optimizations and/or high-GC protocol parameters.