Tapestri Single-Cell DNA and DNA + Protein Sequencing User Guide **Workflow Changes**

	Chapter	Step	V2	V3
0	Firmware	Instrument Firmware		Update/ensure **V3 compatible firmware**
1	Prepare Cell Suspension	Dilute Cell Suspension	**3,000 - 4,000** cells/µL	**2,800 - 3,200** cells/µL
	Encapsulate Cells	Reagent Retrieval		Equilibrate Encapsulation Oil to RT **from 4°C storage**
2		Prepare Lysis Mix	Prepare 100 µL Lysis Mix, **use 90 µL**	Prepare 70.1 µL Lysis Mix, **use 60 µL**
		Post Encapsulation	**Remove most oil** leaving <5 µL only	Remove oil until **total volume (emulsion + oil) = 100 µL**
3	Lyse & Digest Cells	No Change		
4	Barcode Cells	Reagent Retrieval	Equilibrate Barcoding Beads to RT **from 4°C storage**	Equilibrate Barcoding Beads to RT **from -20°C storage** Equilibrate Barcoding Oil to RT **from 4°C storage**
		Barcoding Bead (BC) Preparation	**Use Barcoding Beads as is**	**Add 67 µL of Barcode Mix to Barcoding Beads** prior to vortexing
		Barcoding	Load **200 µL Barcoding Beads** into reservoir 7	Load **250 µL Barcoding Beads** into reservoir 7
			Load **250 µL of Barcode Mix** into reservoir 8	Load **200 µL of Barcode Mix** into reservoir 8
		Post Barcoding	Remove **up to 120 µL oil** from each tube	Remove oil until the **total volume (emulsion + oil) = 100 µL** from each tube
5	Targeted Amplification	No Change		
6	Cleanup PCR Products	Digest PCR Product	**Pool tubes 1-4 and 5-8 into separate tubes** and add 20 µL DNA Clean up Buffer and 12 µL Clean up Enzyme Incubate in **four 0.2 mL tubes**	**Pool tubes 1-8 into one tube** and add 40 µL DNA Clean up Buffer and 24 µL Clean up Enzyme Incubate in **one 1.5 mL tube**
		Clean up PCR product		After enzymatic digest, spin down tube and **transfer supernatant leaving behind any visible pellet**
		Clean up PCR product	Perform AMPure XP bead cleanup in **2 tubes**	Perform AMPure XP bead cleanup in a **single tube**
		DNA Library Cleanup I	Perform one 0.72x (DNA) or 0.70x (DNA+Protein) AMPure XP cleanup	Perform one 0.72x (DNA) or 0.70x (DNA+Protein) AMPure XP cleanup **followed by a second 0.76x AMPure XP cleanup**
7	Library PCR	DNA Library Cleanup II	Perform **one 0.69x AMPure XP** cleanup	Perform one 0.69x AMPure XP cleanup **followed by a second 0.72x AMPure XP cleanup**
8	Quantify and Normalize Library	No Change		
9	Sequence Library	Required reads	**~5,000 cells expected**	**~11,000 cells expected**