Supplementary Figures

Figure SI1

Figure SI1A



Figure SI1B

Patient ID	Parsortix DEPArray count (vol. of blood analysed)	Adjusted Parsortix DEPArray count (corrected for DEP dead volume)	Final Parsortix sample count (extrapolated to 7.5 mL)
Patient 1	344 (2.5 mL)	491	1474
Patient 2	103 (2.125 mL)	147	519
Patient 3	112 (2.125 mL)	160	565
Patient 4	59 (4.25 mL)	84	149
Patient 5	88 (4.25 mL)	126	222
Patient 6	56 (2.125 mL)	80	282
Patient 7	22 (4.25 mL)	31	55
Patient 8	11 (2.125 mL)	16	55
Patient 9	13 (2.125 mL)	19	66
Patient 10	6 (2.125 mL)	9	30
Patient 11	12 (2.125 mL)	17	61
Patient 12	4 (2.125 mL)	6	20

Figure SI1C

Patient ID	CTC counts (adjusted to 7.5 mL of blood)		
Patientib	CellSearch	Parsortix	
Patient 1	1167	1474	
Patient 2	3780	519	
Patient 3	1	565	
Patient 4	89	149	
Patient 5	43	222	
Patient 6	174	282	
Patient 7	27	55	
Patient 8	0	55	
Patient 9	0	66	
Patient 10	4	30	
Patient 11	6	61	
Patient 12	1	20	

Figure SI1. Parsortix clinical CTC count calculations. (A) Workflow for assessing the CTC count appropriate for 7.5 mL of patient blood. The CTC counts from the DEPArray (cells which were CK positive, CD45 negative with good nuclear staining) were first extrapolated to account for the 30% dead volume of the DEPArray, after which they were extrapolated to account for 7.5 mL of sample (the volume analysed using the CellSearch). (B) CTC counts for all patient samples analysed at every stage of the calculation. Fractions accounting for different volumes of blood were analysed, the final calculation normalised all samples to 7.5 mL. (C) Comparison of CTC counts for 7.5 mL of blood from paired samples analysed by CellSearch and using the Parsortix workflow.

Figure SI2

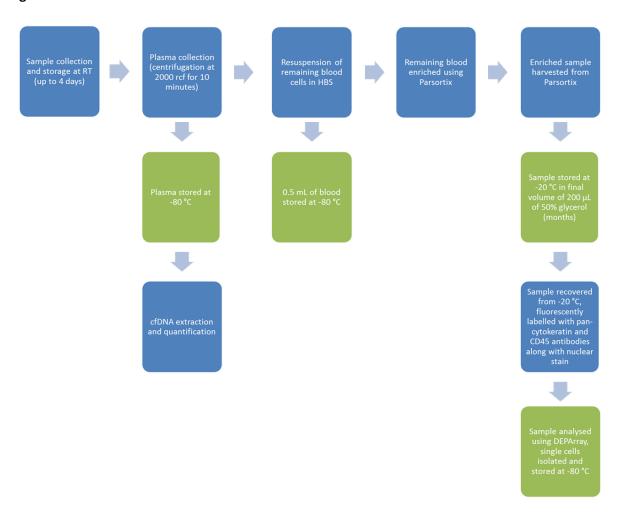


Figure SI2. Clinical sample processing workflow. Steps involving sample manipulation are shown in blue, steps involving storage of samples derived from the original blood sample are shown in green.

Figure SI3

Figure SI3A

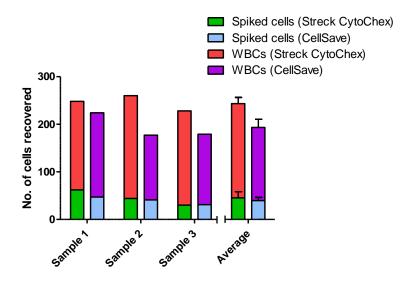


Figure SI3B

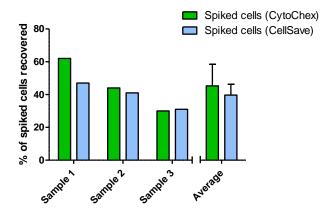


Figure SI3C

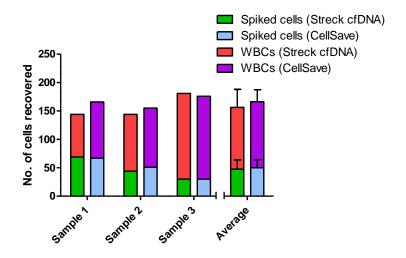


Figure SI3D

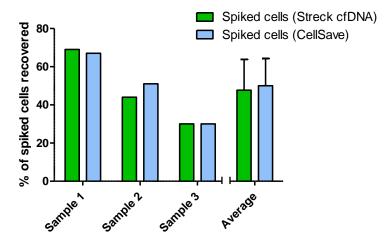


Figure SI3E

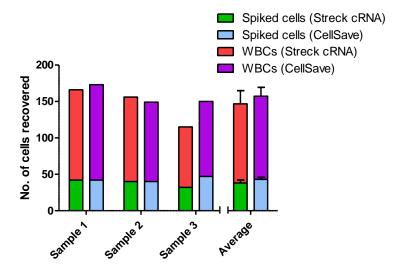


Figure SI3F

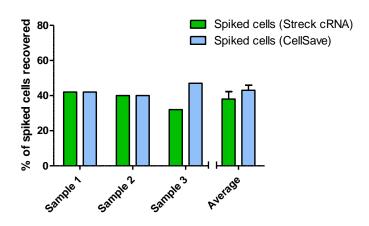


Figure SI3. Comparison of CellSave preservative with Streck CytoChex, Cell-Free DNA and Cell-Free RNA preservatives. Paired samples of 2 mL of healthy normal volunteer blood preserved in either CellSave or one of the three Streck preservatives were spiked with 100 H2009 cells pre-labelled with CellTracker Green and enriched using the double chip protocol. (A) Numbers of spiked cells and white blood cells recovered from paired samples preserved in CellSave and Streck CytoChex. (B) Spiked cell recovery percentages for samples preserved in CellSave and Streck CytoChex. (C) Numbers of spiked cells and white blood cells recovered from paired samples preserved in CellSave and Streck Cell-Free DNA BCT. (D) Spiked cell recovery percentages for samples preserved in CellSave and Streck Cell-Free DNA BCT. (E) Numbers of spiked cells and white blood cells recovered from paired samples preserved in CellSave and Streck Cell-Free RNA BCT. (F) Spiked cell recovery percentages for samples preserved in CellSave and Streck Cell-Free RNA BCT.