

Analytical performance of the FDA-cleared Parsortix[®] PC1 system

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ABSTRACT

Introduction: The Parsortix[®] PC1 system, Food and Drug Administration (FDA) cleared for use in metastatic breast cancer (MBC) patients, is an epitope-independent microfluidic device for the capture and harvest of circulating tumor cells from whole blood based on cell size and deformability. This report details the analytical characterization of linearity, detection limit, precision, and reproducibility for this device.

Methods: System performance was determined using K₂-EDTA blood samples collected from self-declared healthy female volunteers (HVs) and MBC patients spiked with pre-labeled cultured breast cancer cell lines (SKBR3, MCF7, or Hs578T). Samples were processed on Parsortix[®] PC1 systems and captured cells were harvested and enumerated.

Results: The system captured and harvested live SKBR3, MCF7, and Hs578T cells and fixed SKBR3 cells linearly between 2 and ~100 cells, with average harvest rates of 69%, 73%, 79%, and 90%, respectively. To harvest ≥1 cell ≥95% of the time, the system required 3, 5 or 4 live SKBR3, MCF7 or Hs578T cells, respectively. Average harvest rates from precision studies using 5, 10, and ~50 live cells spiked into blood for each cell line ranged from 63.5% to 76.2%, with repeatability and reproducibility percent coefficient of variation (%CV) estimates ranging from 12.3% to 32.4% and 13.3% to 34.1%, respectively. Average harvest rates using ~20 fixed SKBR3 cells spiked into HV and MBC patient blood samples were 75.0% ± 16.1% (%CV = 22.3%) and 68.4% ± 14.3% (%CV = 21.1%), respectively.

Conclusions: These evaluations demonstrate the Parsortix[®] PC1 system linearly and reproducibly harvests tumor cells from blood over a range of 1 to ~100 cells.

Keywords: Blood, Breast cancer, Circulating tumor cells, Liquid biopsy, Microfluidic devices, Neoplastic cells

Introduction

There is increasing interest in using liquid biopsy in the clinical setting as a tool for personalized medicine through

the detection and characterization of cells that could seed metastasis, for longitudinal monitoring of cancer patients in real time, identifying treatment regimens or response and/or minimal residual disease (MRD). A liquid biopsy is ideal for these purposes due to the minimally invasive and repeatable nature of a blood draw, in contrast to traditional tissue biopsies (1-3).

Circulating tumor cells (CTCs) are cancer cells that have intravasated into the bloodstream or lymphatic system from primary and/or metastatic tumors (4,5). CTCs released from primary tumors into the bloodstream can travel to distant sites and form secondary tumors, an integral part of the metastatic process. Epithelial-mesenchymal transition (EMT) is a crucial process enabling CTC detachment, intravasation *into* the circulation, extravasation *from* the circulation, and, ultimately, metastases formation (1,5-7). Cancer cells circulate both as single CTCs and CTC clusters, with CTC clusters showing an increased metastatic potential (5,8). Isolating and

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characterizing the cells that have the potential to metastasize (i.e., CTCs) is vitally important, considering that ~90% of cancer deaths are due to metastasis (9).

Isolation and interrogation of CTCs has been challenging due to the rarity of these cells in the bloodstream, with approximately one cell in 10^5 – 10^6 peripheral blood mononuclear cells (PBMCs) (4,5,10). Yet it has been documented that the detection of CTCs is a clinically relevant predictor of disease progression and overall survival in patients with metastatic breast cancer (MBC) (11-16). A recent study using the Parsortix® system to obtain CTCs from blood reported a high concordance in predictive biomarker expression between the CTCs and tissue from the metastases, demonstrating that analysis of CTCs may serve as a surrogate sample for evaluation of biomarkers for breast cancer metastases (17).

Many technologies have been developed for CTC isolation, including biological and physical-based methods (7,18,19). Biological-based methods isolate CTCs based on phenotype and rely on antibodies to select cells of interest. Many of these, including the CellSearch® System, isolate CTCs based on expression of the epithelial cell adhesion molecule (EpCAM), which is expressed by a subset of CTCs. CTCs released into the bloodstream are often either undergoing or have undergone EMT, which involves upregulation of mesenchymal gene expression and downregulation of epithelial gene expression (20). This results in failure of EpCAM-based methods to efficiently isolate mesenchymal and/or EMTing CTCs, the CTC phenotype that may be more representative of cells capable of forming distant metastases (19). In addition, the use of antibodies for capture could impact downstream analysis (18,21) of the captured CTCs. These limitations identify a critical need for physical-based, or epitope-independent, CTC isolation methods enabling the capture and subsequent characterization of broader CTC populations.

The Parsortix® PC1 system is a semi-automated, epitope-independent microfluidic device that captures and subsequently harvests rare cells (including CTCs) from whole blood based on their size and deformability (22-29). This physical-based method of CTC isolation is not reliant on specific antigen expression for capture and enables enrichment of both epithelial and mesenchymal cells (22,24,25,29). The system also harvests the captured cells from the microfluidic device into a small volume of buffer (~210 μ L) for subsequent user-defined evaluation.

The studies reported here were designed to evaluate the analytical performance of the Parsortix® PC1 system for the capture and harvest of breast cancer cells, including assessment of linearity, limit of detection (LoD), precision, and reproducibility, using whole blood samples spiked with live or fixed cultured breast cancer cell lines (SKBR3, MCF7, or Hs578T). The breast cancer cell lines chosen for spiking are representative of the heterogeneity observed in breast cancer and breast cancer CTCs (30-33). The analytical performance data generated herein plus the previously published clinical performance data (34) were included in the de novo request for the classification of the Parsortix® PC1 system (DEN200062) as a Class II medical device, which the Food and Drug Administration (FDA) granted in May 2022 ([Online](#)).

Methods

Blood sample collection

Blood was collected via venipuncture from self-declared healthy female volunteers (HVs) and via either a venipuncture or an existing port from MBC patients into 10 mL K_2 -ethylenediaminetetraacetic acid (EDTA) tubes (Becton Dickinson). A minimum of 7.5 mL of blood per tube, up to a maximum of 80 mL of blood per donation, was obtained from each HV. A minimum of 5 mL of blood per tube was obtained from each MBC patient. The HVs were self-declared healthy, nonpregnant women aged ≥ 18 years with no known current illness or other serious health condition. MBC patients were women with documented evidence of MBC (as determined by any means). All women (HVs and MBC patients) gave written informed consent prior to collection of the blood samples under protocols that were reviewed and approved by the London-Surrey Borders Research Ethics Committee or the University of Rochester Research Study Review Board.

Cell lines

The breast cancer cell lines SKBR3, MCF7, and Hs578T were obtained from the American Type Culture Collection (ATCC) and were chosen because they are representative of CTCs that would be found in the blood of patients with MBC based on their immunotype, phenotype, morphology, and size (Tab. I) (30-33,35-38). Cell line maintenance is detailed in the Supplementary Materials.

TABLE I - Comparison of phenotype, immunophenotype, morphology, and size for SKBR3, MCF7, Hs578T, and CTCs

	SKBR3	MCF7	Hs578T	MBC patient-derived CTCs
Breast cancer subtype	HER2 enriched	Luminal A	Basal/triple negative	All subtypes
Phenotype	Epithelial	Epithelial	Mesenchymal	Epithelial, EMT, and mesenchymal
Receptor status	ER/PR- and HER2+	ER/PR+ and HER2-	ER/PR- and HER2-	ER/PR+/- and HER2-/+
Morphology	Grape-like	Mass	Stellate	Highly polymorphic
Size	15-17 μ m (microscope and counter pipette) (Coumans et al (35))	16.5 μ m (counter pipette) (Coumans et al (35))	No data	13.1 μ m (Coumans et al (35)) 11.79 (4.51-33.11) μ m (Zhao et al (36)) 13.1 (12-25) μ m (Hao et al (37)) 11 (6-16) μ m ² (Coumans et al (38))

CTC = circulating tumor cell; EMT = epithelial-mesenchymal transition; MBC = metastatic breast cancer.

For each study, using the abovementioned cell lines, live or fixed cells (fixed using 4% formaldehyde [Sigma Aldrich]) were fluorescently labeled with CellTracker™ Green 5-chloromethylfluorescein diacetate (CMFDA; Life Technologies) as per the manufacturer's instructions. The labeled cell lines were spiked into freshly collected blood samples as detailed below.

Blood sample preparation

For linearity, LoD, and live cell precision studies, spike levels of ≤ 100 cells were directly spiked, as a single droplet, onto the side of empty 12 mL polystyrene tubes. The number of cells in the droplet was counted by two independent operators using a Leica fluorescence microscope. When an accepted cell count within the droplet (see Tab. SI) was confirmed by two independent operators, 7.5 mL of blood was added to the tube, ensuring the blood flowed over the droplet containing the spiked cells. Spike levels of >100 cells were spiked directly into blood samples from cell dilutions prepared in phosphate-buffered saline (PBS) containing 1% bovine serum albumin (BSA; w/v) and 2 mM EDTA (BEP) (total volume of spike ≤ 5 μ L). The numbers of cells spiked were calculated by spiking the same volume of cell dilution into six wells of a 96-well plate, each well containing 200 μ L of $1\times$ PBS (control plate), and either counting manually or using the Leica fluorescent microscope software and taking the mean of the six control spikes.

For precision and reproducibility studies using ~ 20 fixed, prelabeled SKBR3 cells, microfuge tubes containing known numbers of fixed, prelabeled SKBR3 cells (range between 17 and 25 cells) in ~ 180 μ L of BEP were prepared ("Precision Tubes"). For spiking, the contents of a Precision Tube were pipetted into either 2.5 mL of PBS or a whole blood sample (study dependent), the tube was rinsed with 80 μ L of additional PBS, and the 80 μ L of additional PBS was pipetted into the PBS or whole blood sample.

ANGLE R&D Laboratory located in Guildford, UK, prepared and processed the samples unless stated otherwise.

Blood sample processing

All blood samples were processed within 8 hours of collection using Parsortix® PC1 systems and GEN3 6.5 μ m cell separation cassettes at a constant pressure of 99 mbar (22). Captured cells were harvested from the cassettes directly into single wells on 96-well plates or onto polytetrafluoroethylene ("PTFE") printed slides with 21 hydrophobic droplet spots for subsequent enumeration using a fluorescence microscope. The number of cells in each harvest was counted by a minimum of two independent operators.

Linearity evaluation

Linearity was determined for each cell line individually by preparing HV blood samples spiked with 2 to ~ 100 live or fixed (SKBR3 only), prelabeled cells (Tab. SI). Data were obtained from a minimum of 10 HVs per cell line and eight different spike levels, with each HV providing data for all spike levels. Additional blood samples were prepared by spiking ~ 125 , ~ 250 , ~ 500 , and ~ 1000 live, prelabeled SKBR3 cells into 7.5 mL aliquots of blood. Data for the higher cell spike

levels were obtained from a minimum of 10 HVs for each spike level, with each HV providing data for all spike levels. The linearity studies were designed in accordance with the Clinical and Laboratory Standards Institute (CLSI; formerly NCCLS) guideline *EP06-A: Evaluation of Linearity of Quantitative Measurement Procedures: A Statistical Approach*.

Limit of blank and LoD evaluation

Limit of blank (LoB) of the system was determined using unspiked HV blood samples. LoD was determined for each cell line individually by preparing HV blood samples directly spiked with 1, 2, 3, 4, or 5 live, prelabeled cells. Data were obtained from a minimum of 60 samples for each spike level and cell line, performed in accordance with the CLSI guideline *EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline—Second Edition*.

Precision evaluation

The precision studies were conducted in accordance with the CLSI guideline *EP05-A3: Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline—Third Edition*. Imprecision estimates for the Parsortix® PC1 system were evaluated for the different cell lines and spike levels using all three cell lines directly spiked into 7.5 mL aliquots of HV blood. For each cell line (SKBR3, MCF7, and Hs578T), 7.5 mL aliquots of blood from HVs were spiked with 5, 10, or ~ 50 live, prelabeled cells. For each spike level and cell line, data were obtained from a total of 100 samples, processed on 10 Parsortix® PC1 systems, two runs per system per day over a total of 5 nonconsecutive days. Additional precision studies evaluating fixed versus live cells, different cassette lots, and different laboratories were also conducted, and a description of these studies and their results can be found in the Supplementary Materials.

Reproducibility evaluation

A separate reproducibility study was conducted at the University of Rochester Medical Center (URMC). For this study, ≥ 5 mL of blood was collected from HVs and MBC patients into 10 mL K_2 -EDTA tubes. The blood tubes were spiked with ~ 20 fixed SKBR3 cells using Precision Tubes. Data were collected from a total of 150 spiked samples (76 HVs and 74 MBC patients) processed on four different Parsortix® PC1 systems.

Data analysis

A detailed description of the data analysis methods used for each study can be found in the Supplementary Materials.

Results

Linearity

The harvesting of live, prelabeled SKBR3, MCF7, and Hs578T and fixed SKBR3 cells by the Parsortix® PC1 system



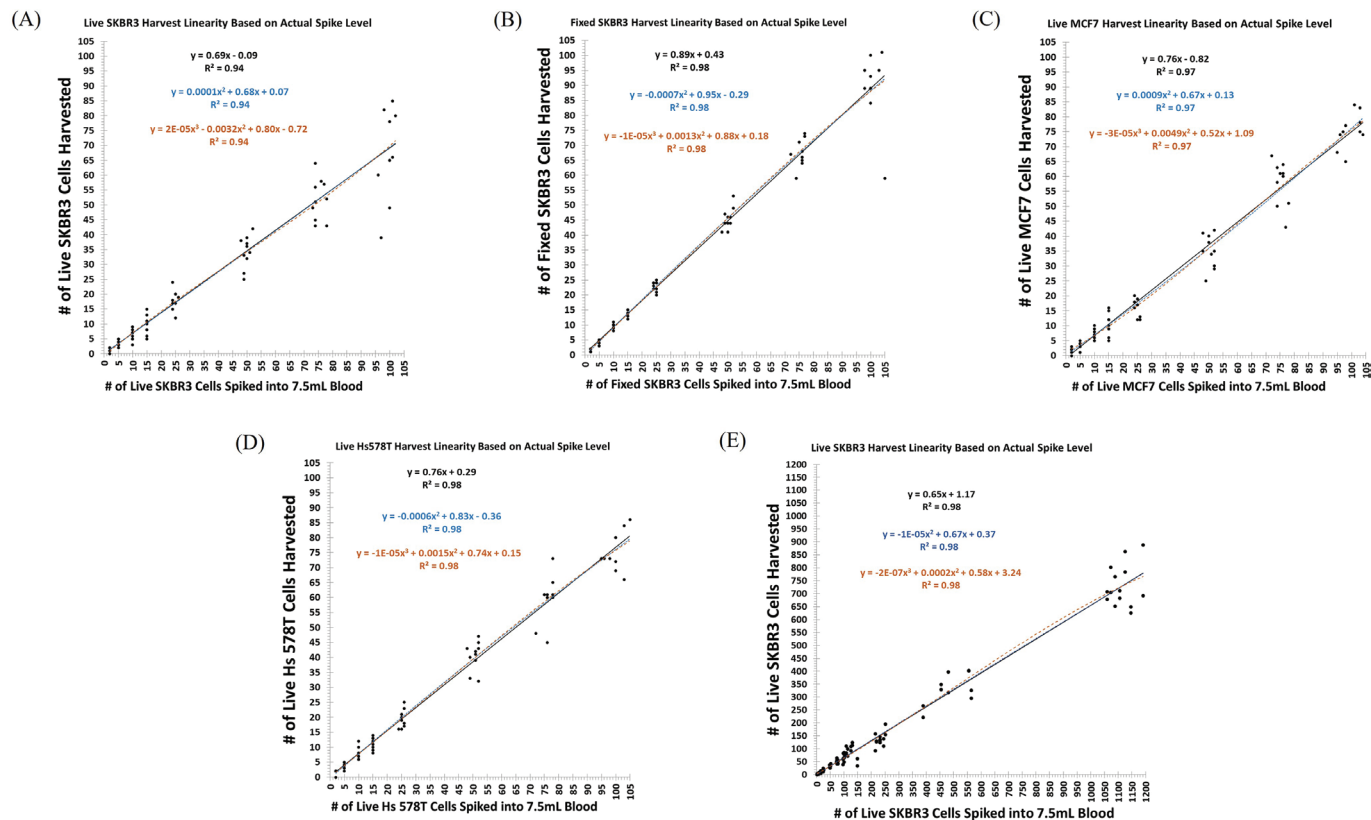


Fig. 1 - Parsortix® PC1 system observed linearity when processing blood samples spiked with a range of cell numbers for live SKBR3, MCF7, and Hs578T cells and fixed SKBR3 cells. Linear (first-order polynomial) regression analysis indicated in black, second-order polynomial regression analysis indicated in blue, and third-order polynomial regression analysis indicated in orange. (A) Live SKBR3 harvest linearity for between 2 and ~100 cells spiked into 7.5 mL of blood, with first-, second-, and third-order polynomial regression analyses shown (34). (B) Fixed SKBR3 harvest linearity for between 2 and ~100 cells spiked into 7.5 mL of blood, with first-, second-, and third-order polynomial regression analyses shown. (C) Live MCF7 harvest linearity for between 2 and ~100 cells spiked into 7.5 mL of blood, with first-, second-, and third-order polynomial regression analyses shown (34). (D) Live Hs578T harvest linearity for between 2 and ~100 cells spiked into 7.5 mL of blood, with first-, second-, and third-order polynomial regression analyses shown (34). (E) Live SKBR3 harvest linearity for between 2 and ~1,000 cells spiked into 7.5 mL of blood, with first-, second-, and third-order polynomial regression analyses shown.

was demonstrated to be linear over the range of 2 to ~100 cells spiked into 7.5 mL of blood (Fig. 1A-D). Additionally, linearity of harvest was shown to extend to at least 1,000 live SKBR3 cells spiked into 7.5 mL of blood (Fig. 1E). The best fit model for all cell lines was the first-order linear regression model (the solid black line).

Over the range of 2 to ~100 *live* SKBR3 cells, the linear regression model had a slope of 0.69, indicating an average harvest rate of ~69% (95% confidence interval [CI] = 65%-73%, $R^2 = 0.94$) (Fig. 1A) (34), whereas over the same range, *fixed* SKBR3 cells resulted in a slope of 0.89, indicating an average harvest rate of ~89% (95% CI = 85%-92%, $R^2 = 0.98$) (Fig. 1B). The data show both an increased variability for the recovery of live SKBR3 cells and better recovery of the fixed SKBR3 cells. This was not unexpected due to the effects of fixation on cultured cells, resulting in reduced deformability of the fixed cells compared to live cells. Over the range of 2 to ~100 live MCF7 cells, the linear regression model had a slope of 0.76, indicating an average harvest rate of ~76% (95% CI = 73%-79%, $R^2 = 0.97$) (Fig. 1C) (34) and over the same range, live Hs578T cells showed similar results with

a slope of 0.76, indicating an average harvest rate of ~76% (95% CI = 74%-79%, $R^2 = 0.98$) (Fig. 1D) (34). Average harvest rates for each cell spike level, along with the standard deviation (SD) and percent coefficient of variation (%CV) for each cell line can be found in Tables SII–SV.

Detection limit (LoB, LoD)

Using a total of 63 unspiked blood samples, the LoB of the Parsortix® PC1 system was determined to be zero cells, as expected. Only one (1.6%) of the 63 unspiked blood samples was found to have a single prelabeled cell harvested. Considering the results from a separate cell carryover study where no carryover of cells was observed after the spiking of >2,000 cultured cells, the observation of a single cell in an unspiked sample is likely the result of a user error (e.g., improper labeling of the sample as a control when it was actually a spiked sample).

The LoD was determined for each cell line separately using a type II error level (β = false negative rate) of <5%, which equated to the spike level at which $\geq 95\%$ of the replicate



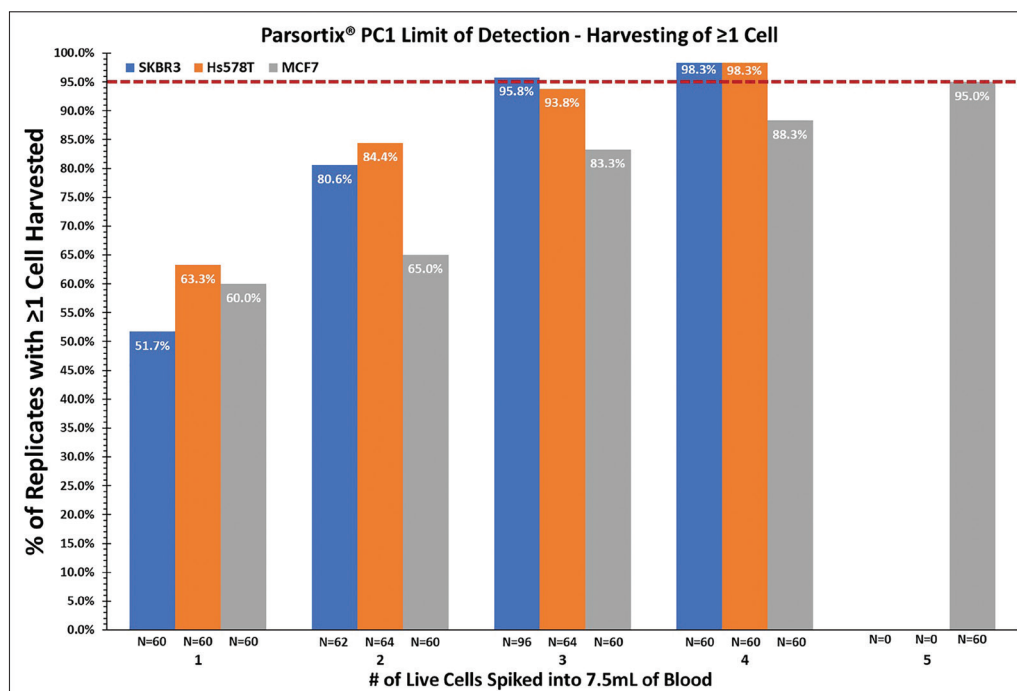


Fig. 2 - Parsortix® PC1 system limit of detection. Percent of replicates with ≥ 1 cell harvested when blood was spiked with 1, 2, 3, 4, or 5 live SKBR3, MCF7, or Hs578T cells. *Note:* For the SKBR3 and Hs578T cell lines, the 5-cell spike level was not evaluated as both of these cell lines showed recoveries of >1 cell at lower cell spike levels.

samples for a given cell line had ≥ 1 cell observed in the harvests (Fig. 2). For the SKBR3 and Hs578T cell lines, spiking of 5 live cells into blood was not performed as the detection limit, as per the specified definition, had already been reached at a cell spike level of fewer than 5 cells.

For live SKBR3 cells, the LoD was 3 cells (95.8% of the replicates spiked with 3 cells had ≥ 1 cell observed in the harvest); for live MCF7 cells, the LoD was 5 cells (95.0% of the replicates spiked with 5 cells had ≥ 1 cell observed in the harvest); and for live Hs578T cells, the LoD was 4 cells (98.3% of the replicates spiked with 4 cells had ≥ 1 cell observed in the harvest).

Precision

Precision studies were performed to assess the ability of the Parsortix® PC1 system to harvest multiple live breast cancer cell lines that are representative of the heterogeneity observed in breast cancer and breast cancer CTCs (30-33) at different spike levels. For each cell line (SKBR3, MCF7, and Hs578T) and spike level, 100 contrived samples consisting of live, prelabeled cells spiked into blood were processed using 10 different Parsortix® PC1 systems over 5 nonconsecutive days in each study. Imprecision estimates in these 5-day precision studies were determined for each cell line at three different spike levels (5, 10, and ~ 50 cells), with the cells being spiked into 7.5 mL of HV blood. The overall average harvest rates for the SKBR3, MCF7, and Hs578T cell lines were 72.0%, 70.9%, and 66.7%, respectively. The average harvest rates for each of the cell lines and cell spike levels ranged from 63.5% to 76.2%, the repeatability (within-run) %CV estimates ranged from 12.3% to 32.4%, and the reproducibility (within-laboratory) %CV estimates ranged from 13.3% to 34.1% (Tab. II). The repeatability and reproducibility %CV estimates

for all of the cell line and cell spike level results combined were 26.3% (95% CI = 25.0%-27.6%) and 26.3% (95% CI = 25.2%-27.8%), respectively, with between-cell-type and between-spike-level %CV estimates of 6.6% (95% CI = 4.0%-10.6%) and 4.6% (95% CI = 1.6%-8.3%), respectively.

Additional precision studies were performed to evaluate the precision of the Parsortix® PC1 system for the capture and harvesting of tumor cells, taking into consideration changes in different variables including live versus fixed cells, separation cassette lots, and laboratory sites. A graphical representation of results from these additional precision studies evaluating live versus fixed cells, different cassette lots, and different laboratories can be seen in Figure 3, with further details provided in the Supplementary Materials.

Reproducibility

The overall combined average percentage of the ~ 20 prelabeled, fixed SKBR3 cells spiked into the blood samples from the 76 evaluable HV subjects and 74 evaluable MBC patients that were harvested was $70.0\% \pm 15.4\%$ (95% CI = 62.3%-76.7%, median = 70.7%). The average percentage of spiked SKBR3 cells harvested in the HV subjects was $72.1\% \pm 16.1\%$ (95% CI = 61.1%-80.9%, median = 75.0%) compared to $67.9\% \pm 14.3\%$ (95% CI = 56.7%-77.4%, median = 68.4%) in the MBC patients (t-test p value = 0.098). Figure 4 shows a scatterplot of the percent harvest results in the HV subjects and MBC patients.

Although the average percent harvest results were not statistically significantly different between the two groups, the average percent harvest results in the MBC patients appeared to be slightly lower. As this was not expected, a possible reason for this was investigated, the results of which are described in the Supplementary Materials

TABLE II - Average harvest rates and imprecision estimates for the Parsortix® PC1 system when harvesting live SKBR3, MCF7, and Hs578T cells spiked into 7.5 mL of blood (2-factor ANOVA models using day and run as the factors)

Cell line	# of cells spiked	N	Mean % harvest	Within-run (Repeatability)		Between run		Between day		Within laboratory	
				SD	CV%	SD	%CV	SD	%CV	SD	%CV
Live SKBR3	5	100	73.6	22.8%	31.0	6.7%	9.1	0.0%	0.0	23.7%	32.2
	10	100	70.4	18.5%	26.3	0.0%	0.0	4.4%	6.3	19.1%	27.1
	~50 (48-52)	100	72.1	13.8%	19.1	0.0%	0.0	0.0%	0.0	13.8%	19.1
	5~50	300	72.0	18.6%	25.8	3.5%	4.9	0.0%	0.0	18.9%	26.3
Live MCF7	5	100	68.0	22.0%	32.4	7.3%	10.7	1.3%	1.9	23.2%	34.1
	10	100	68.4	18.4%	26.9	0.0%	0.0	6.3%	9.2	19.5%	28.5
	~50 (48-52)	100	76.2	11.3%	14.8	0.0%	0.0	3.4%	4.5	11.8%	15.5
	5~50	300	70.9	18.5%	26.1	2.1%	3.0	1.6%	2.3	18.7%	26.4
Live Hs578T	5	100	66.6	19.9%	29.9	5.9%	8.9	3.6%	5.4	21.0%	31.5
	10	100	63.5	17.8%	28.0	0.0%	0.0	7.5%	11.8	19.3%	30.4
	~50 (48-52)	100	70.0	8.6%	12.3	0.0%	0.0	3.4%	4.9	9.3%	13.3
	5~50	300	66.7	16.8%	25.2	2.3%	3.4	3.8%	5.7	17.3%	25.9
Live SKBR3, MCF7, and Hs578T	5	300	69.4	22.6%	32.6	3.6%	5.2	0.0%	0.0	22.8%	32.9
	10	300	67.4	18.6%	27.6	0.0%	0.0	3.8%	5.6	19.0%	28.2
	~50 (48-52)	300	72.8	11.6%	15.9	0.0%	0.0	2.6%	3.6	11.9%	16.3
	5~50	900	69.9	18.4%	26.3	1.4%	2.0	0.0%	0.0	18.4%	26.3

ANOVA = analysis of variance; %CV = percent coefficient of variation; SD = standard deviation.

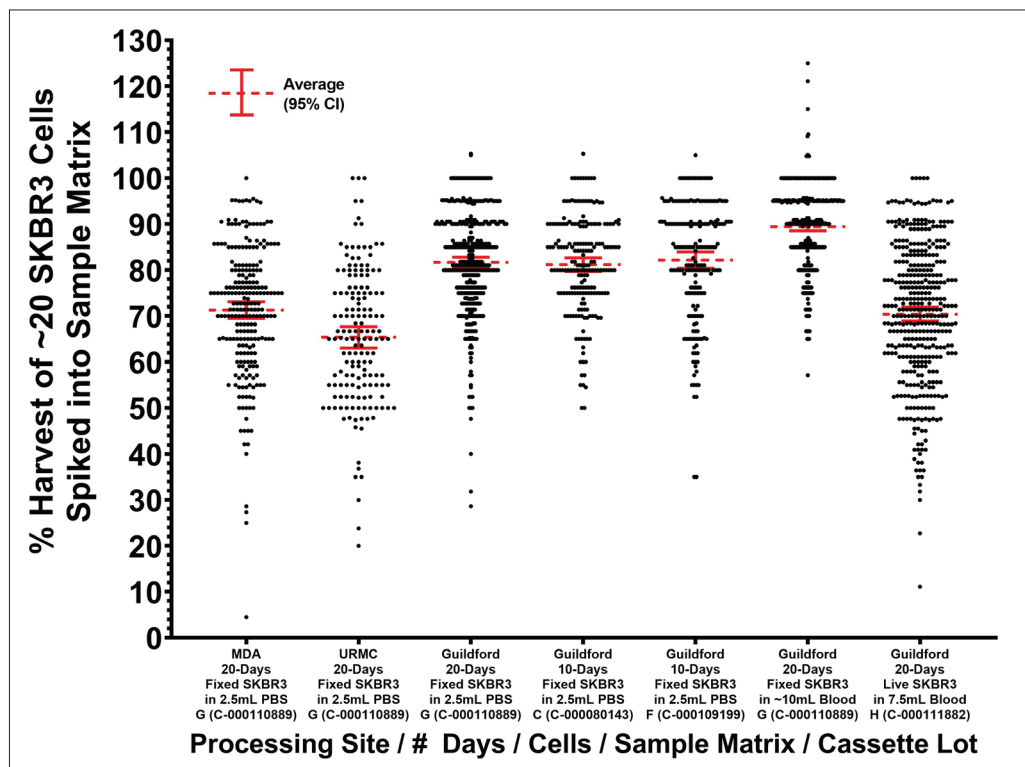


Fig. 3 - Precision of the Parsortix® PC1 system. Percent harvest (average and 95% CI shown) when spiking ~20 fixed SKBR3 cells into 2.5 mL PBS at different laboratory sites (MDA, URM, ANGLE R&D) and using different separation cassette lots (Lot G, Lot C, and Lot F) or spiking ~20 fixed SKBR3 into 10 mL of blood, or ~20 live SKBR3 cells into 7.5 mL of blood. CI = confidence interval; PBS = phosphate-buffered saline.



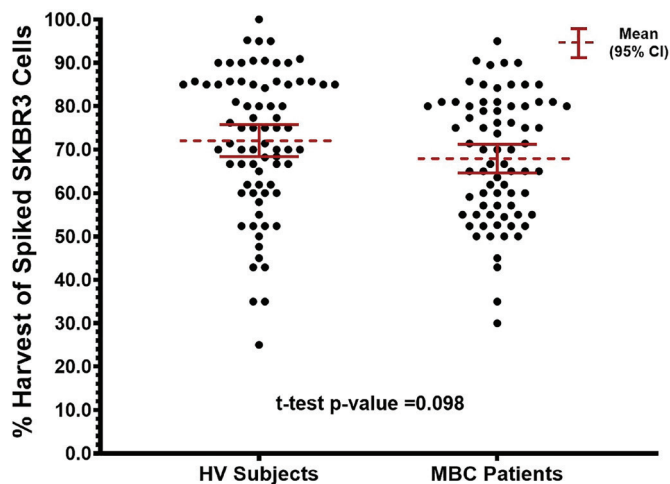


Fig. 4 - Reproducibility of the Parsortix® PC1 system. Percent harvest results from HV subject and MBC patient blood samples (≥ 5 mL of blood from a 10 mL K_2 -EDTA tube) spiked with ~ 20 fixed, pre-labeled SKBR3 cells. EDTA = ethylenediaminetetraacetic acid; HV = healthy female volunteer; MBC = metastatic breast cancer.

(Supplementary Figure 1A-B). This investigation showed significant differences in the average percent harvest results between different batches of fixed SKBR3 cells, providing evidence that the model system used in the analytical studies (i.e., cultured cell lines) contributed to the observed variabilities. This suggests that the actual imprecision of the Parsortix® PC1 system for the capture and harvest of CTCs may be lower than what was observed in these studies.

Conclusions

The analytical performance data show that the Parsortix® PC1 system harvested cultured breast cancer cells in a reproducible and linear fashion, with a minimum of 3 to 5 cells per 7.5 mL of blood required to ensure at least one cell will be harvested most of the time. The average repeatability and reproducibility %CV estimates for all of the live cell lines, chosen because they are representative of the heterogeneity expected to be seen in breast cancer CTCs, and all spike levels (5, 10, and ~ 50 cells) were both 26.3%, with the imprecision estimates decreasing as the number of cells spiked increased. Comparison of live and fixed SKBR3 cells showed that imprecision in the percentage of cells harvested by the system was higher when using live breast cancer cells, which was expected due to the increased deformability of live cells compared to more rigid fixed cells. This increased variability in harvest rate was also observed in the linearity data when comparing live and fixed breast cancer cells.

The analytical performance data reported here plus the previously published clinical performance data (34) were included in the submission for the de novo request for classification of the Parsortix® PC1 system (DEN200062) as a Class II medical device, which was granted by the FDA in May 2022 ([Online](#)). The Parsortix® PC1 system is the first FDA-cleared medical device for the capture and harvest of CTCs from the blood of MBC patients for subsequent, user-validated analysis.

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