

**Supplementary Figure 1. Enumeration of CTC subtypes from Study A and Study B.**

**(A and B ) Study A: Presence, sensitivity and specificity of Pathologically definable CTCs (PDCTCs) or Epithelial-Mesenchymal transition CTCs (EMT CTCs) in patients with known**

**invasive breast cancer versus healthy control volunteers.** PDCTCs were found in 31 of 41 patients

with known invasive breast cancer, but in none of the 16 healthy controls. Sensitivity: 76% (CI95% 60-

88%) Specificity: 100% (CI95% 79-100%) PPV: 100% (CI95% 89-100%) NPV: 62% (CI95% 41-80%).

EMT CTCs were found in 19 of 41 patients with known invasive breast cancer, but in none of the 16

healthy controls. Sensitivity: 54% (CI95% 37-69) Specificity: 100% (CI95% 79-100%) PPV: 100%

(CI95% 85-100%) NPV: 46% (CI95% 29-63%).

**(C and D) Study B: Presence, sensitivity and specificity of PCTCs or EMT CTCs in patients with positive mammograms/CBEs compared to standard pathological assessment .** PDCTCs were not

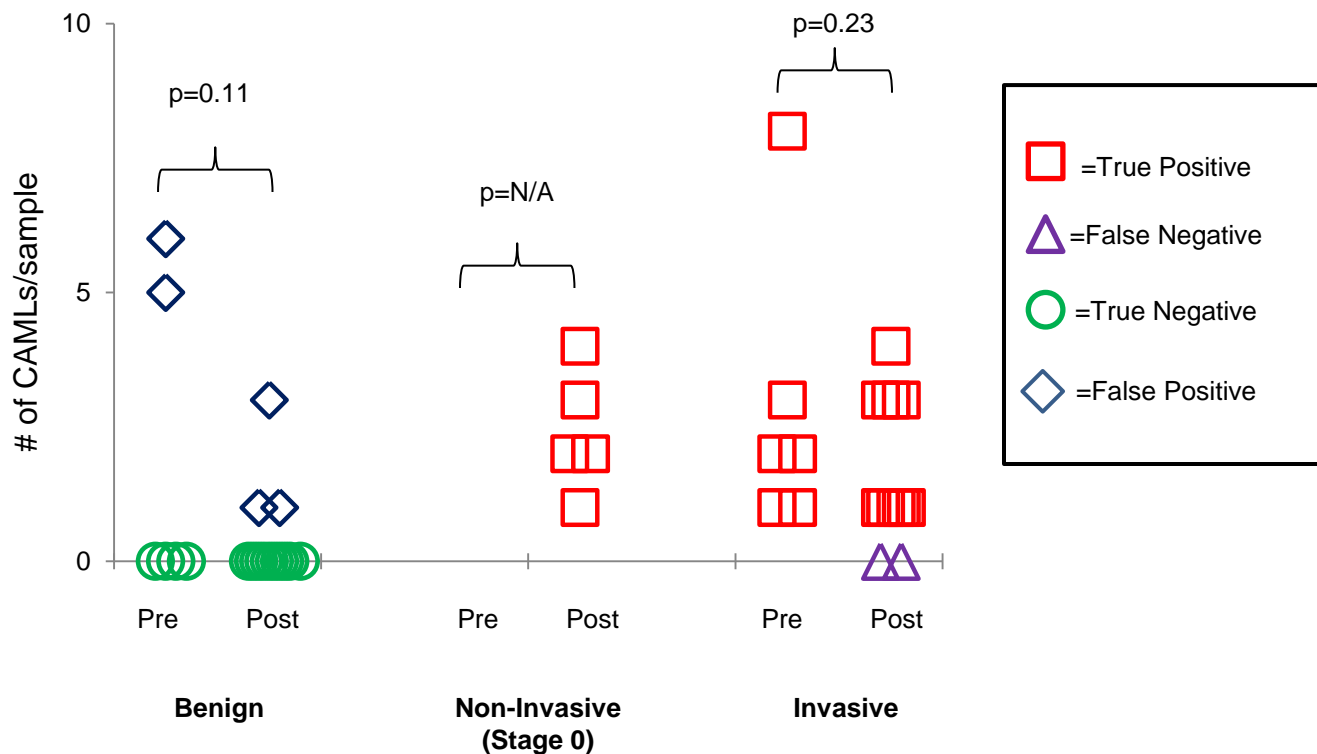
found in any patient samples from Study B. EMT CTCs were found in 40% (n=2/5) of patients with non-

invasive disease, in 18% (n=3/17) of patients with invasive disease, and 37% (n=7/19) of patients with

benign conditions. Comparing invasive breast cancer (n=17) to benign conditions (n=19). Sensitivity:

18% (CI95% 4-43%) Specificity: 63% (CI95% 38-84%) PPV: 30% (CI95% 7-65%) NPV: 46% (CI95%

27-67%).



**Supplementary Figure 2. CAML enumeration from blood samples taken pre or post biopsy.** To account for the possibility that presence of CAMLs may be a residual artifact from manipulation caused by the biopsy, thirteen samples were taken prior to biopsy and twenty nine samples taken post biopsy. There were no samples pre-biopsy and 5 post-biopsy in the non-Invasive group; 6 pre-biopsy and 13 post-biopsy in the benign group; and 6 pre-biopsy and 11 post-biopsy in the Invasive group. An unpaired student's T-test was calculated for each grouping, pre versus post. It was determined that there was no significant difference between pre or post biopsy ( $p>0.05$ ) in either the benign or invasive groupings.

**Supplementary Table 1: Cancer patient characteristics from Study A: Patients with known diagnosed breast cancer**

		Number of patients	% CAML Positive (median)
Stage	1	-	
	2	2	100 (44.5)
	3	13	85 (6)
	4	26	96 (9.5)
ER/PR*	Positive	16	94 (7.5)
	Negative	20	95 (9.5)
	*Unknown	5	80 (1)
HER2*	Positive	12	92 (29.5)
	Negative	24	96 (6.5)
	*Unknown	5	80 (1)
Histology	Ductal	17	94 (3)
	Lobular	3	100 (5)
	†Other	21	90 (22)
Treatment	On therapy	28	96 (25.5)
	No therapy	13	85 (2)

\* Tissue unavailable for subtyping

† Tissue unavailable for histology assessment, and/or patients with unspecified metastatic breast cancer, and/or patients with cancers other than IDC or ILC.

**Supplementary Table 2: Cancer patient characteristics from Study B: patients with invasive breast cancer**

		Number of patients	% CAML Positive (median)
Stage	1	4	50 (0.5)
	2	10	100 (2)
	3	1	100 (4)
	NS	2	100 (2)
Nodal Status	N0	6	84 (1)
	N1	8	88 (2)
	Nx	3	100 (1)
Histologic Grade	1	2	100 (2.5)
	2	11	82 (1)
	3	4	100 (2.5)
ER	Positive	14	86 (1)
	Negative	3	100 (3)
HER2	Positive	3	100 (1)
	Negative	13	85 (1.5)
Histology	Ductal	14	86 (1.5)
	Lobular	3	100 (1)
Lymphovascular Invasion			
	Present	5	80 (1)
	Absent	12	92 (1.5)

**Supplementary Table 3: Summary overview of Sensitivity, Specificity, AUC, and 95% confidence intervals for Both Study groups.**

<b>Study A</b>	<b>Sensitivity (CI95%)</b>	<b>Specificity (CI95%)</b>	<b>AUC (CI95%)</b>	<b>PPV (CI95%)</b>	<b>NPV (CI95%)</b>
Invasive Breast Cancer vs Healthy Control	93% (80-99%)	100% (79-100%)	0.96 (0.91-1.00)	100% (91-100%)	84% (60-97%)
<b>Study B</b>	<b>Sensitivity (CI95%)</b>	<b>Specificity (CI95%)</b>	<b>AUC (CI95%)</b>	<b>PPV (CI95%)</b>	<b>NPV (CI95%)</b>
Invasive Breast Cancer vs Benign conditions	88% (64-99%)	74% (49-91%)	0.78 (0.63-0.92)	75% (51-91%)	88% (62-99%).