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Original Article

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Circulating Tumor Cells Prediction in Hormone Receptor Positive HER2-Negative Advanced Breast Cancer: A Retrospective Analysis of the MONARCH 2 Trial

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Abstract

Background: The MONARCH 2 trial (NCT02107703) showed the efficacy of abemaciclib, a cyclin-dependent kinase 4 & 6 inhibitor (CDK4/6i), in combination with fulvestrant for hormone receptor-positive, HER2-negative metastatic breast cancer (MBC). The aim of this analysis was to explore the prediction of circulating tumor cells (CTCs) stratification using machine learning for hypothesis generation of biomarker-driven clinical trials.

Patients and Methods: Predicted CTCs were computed in the MONARCH 2 trial through a K nearest neighbor (KNN) classifier trained on a dataset comprising 2436 patients with MBC. Patients were categorized into predicted Stage $IV_{aggressive}$ (pStage $IV_{aggressive}$, ≥5 predicted CTCs) or predicted Stage $IV_{indolent}$ (pStage $IV_{indolent}$, <5 predicted CTCs). Prognosis was tested in terms of progression-free-survival (PFS) and overall survival (OS) through Cox regression.

Results: Patients classified as predicted pStage $IV_{aggressive}$ and predicted pStage Stage $IV_{indolent}$ were, respectively, 183 (28%) and 461 (72%). After multivariable Cox regression, predicted CTCs were confirmed as independently associated with prognosis in terms of OS, together with ECOG performance status, liver involvement, bone-only disease, and treatment arm. Patients in the pStage Stage $IV_{indolent}$ subgroup treated with abemaciclib experienced the best prognosis both in terms of PFS and OS. The treatment effect of abemaciclib on OS was then explored through subgroup analysis, showing a consistent benefit across all subgroups.

Conclusion: This study is the first analysis of CTCs modeling for stage IV disease stratification. These results show the need to expand biomarker profiling in combination with CTCs stratification for improved biomarker-driven drug development.

Key words: circulating tumor cell; biomarker; drug development; stage IV; breast cancer.

Implications for Practice

This study presents a first-time analysis of clinical outcomes using predicted modeling of circulating tumor cells (CTCs) to stratify stage IV disease. The results suggest the need to broaden the evaluation of resistance biomarkers in conjunction with CTC stratification for improved biomarker-driven drug development and personalized treatment. As a matter of fact, CTC enumeration can enhance the optimization of clinical trial resources by targeting the aggressive subgroup of stage IV disease, leading to a higher probability of events and requiring a smaller sample size and shorter follow-up in trial design.

Background

Metastatic breast cancer occurs in approximately 20% of patients with early breast cancer (BC) history and in 6%-10% of newly diagnosed BC cases. Different disease subtypes account, at least in part, for the variability in overall survival (OS) which can range from a few months to many years.[1,2] Notwithstanding the recent advances in circulating biomarkers, the most established predictive factors in advanced BC (ABC) are still the expression of hormone receptors and the HER2 status. On the other hand, although being recognized as an independent prognostic marker, the role of circulating tumor cells (CTCs) in supporting clinical management of ABC is still not well defined.[3-5]

The phase III STIC CTC trial randomly assigned 761 ABC patients to either a clinically driven or a CTC-driven treatment based on the established 5 CTC/7.5 mL cutoff (Stage

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$IV_{aggressive}$ vs. Stage $IV_{indolent}$).[4,6] Patients classified as high risk received chemotherapy, whereas those classified as low risk received endocrine therapy (ET). PFS was not inferior in the CTC-driven strategy compared to the clinically driven one, while Stage $IV_{aggressive}$ patients who were clinically defined as low-risk had a significantly longer PFS when treated with chemotherapy compared to those treated with ET.

Although promising as a new potential application of CTC enumeration in supporting clinical decision making, these findings focus on an old question, as first-line chemotherapy is no longer an option of choice outside specific scenarios such as visceral crisis. As a matter of fact, cyclin dependent kinase 4 & 6 inhibitor (CDK4/6i) associated with ET are the mainstay treatment in hormone receptor (HR)-positive, HER2-negative ABC and, with this respect, the MONARCH 2 study showed the efficacy of abemaciclib and fulvestrant in patients previously treated with ET.[7,8]

We previously showed the feasibility of predicting CTCs stratification through a K nearest neighbor (KNN) machine learning classifier, proofing the concept of an in silico simulation for hypothesis generation regarding different levels of treatment intensity in subpopulation with different risk profiles.[4,9]

The aim of this hypothesis-generating analysis was to assess the potential utility of CTCs stratification in HR-positive HER2-negative ABC treated with CDK4/6i utilizing a dataset from a randomized phase III study.

Methods

Study Population and Machine Learning Classifier

Predicted CTCs were computed in the MONARCH 2 (NCT02107703) randomized, phase III trial through a K-nearest neighbor (KNN) machine learning algorithm. MONARCH 2 enrolled patients with HR positive HER2 negative ABC irrespectively of menopausal status who had disease progression following ET. Patients received abemaciclib (150 mg) or placebo twice daily during each 28-day cycle plus 500 mg fulvestrant. Treatment continued until progressive disease, death, or discontinuation for any other reason. Patients with more than one ET or any prior chemotherapy for ABC were excluded. Further trial details and eligibility criteria were previously published.[7,8]

KNN is a supervised machine learning algorithm that can solve classification as well as regression problems. The model was previously trained on a pooled dataset of 2436 ABC patients from the European Pooled Analysis Consortium (EPAC) and the MD Anderson Cancer Center to identify patients likely to have CTCs ≥5/7.5 mL blood (Stage $IV_{aggressive}$ vs. Stage $IV_{indolent}$).[4,6]

The model was trained based on estrogen receptor status (ER) (positive vs. negative), progesterone receptor status (PR) (positive vs. negative), HER2 status (positive vs. negative), treatment line (continuous variable), bone and liver involvement (yes vs. no). Patients with all the necessary features (2248) were then 3:1 randomly assigned to a training set (1687) and a validation set (561).[9]

The classifier had a 65.1% accuracy and its prognostic impact resulted in a hazard ratio (HR) of 1.89 for predicted Stage $IV_{aggressive}$ (pStage $IV_{aggressive}$) vs pStage $IV_{indolent}$ ($P < .001$), similar to patients with actual CTCs enumeration (HR 2.76; $P < .001$).[9] The model further stratified clinical subgroups usually considered prognostically homogeneous such as patients with bone-only or liver metastases.[9] The classifier's performance was moreover tested on an independent retrospective database comprising 446 consecutive HR-positive HER2-negative MBC patients.[9]

The model was built using R (The R foundation for Statistical Computing. version 4.1.0) and the "caret" package.[9,10]

The study was approved by the ethical and local institutional review boards for the clinical trial sites, and it was carried out in accordance with Good Clinical Practice guidelines and the Helsinki Declaration. Before enrolling, all patients provided written informed consent.

Statistical Analysis

Categorical variables were reported as frequency distribution, whereas continuous variables were described through median and interquartile range (IQR).

The tested prognostic factors (ie, ECOG performance status [PS], tumor grade, progesterone receptor status, liver and bone-only involvement) were previously identified in the MONARCH 2 and 3 cohorts.[11]

The MONARCH 2 study's definition of overall survival (OS) was the interval from randomization to death from any cause or the last follow-up date. Similarly, progression-free survival (PFS) was defined as the time from randomization to disease progression (according to RECIST criteria), death from any cause, or date of last follow-up. Patients without an endpoint event at the last follow-up visit were censored. Survival was represented by a Kaplan-Meier estimator plot and examined using the log-rank test and univariable and multivariable Cox regression models. The differential impact of abemaciclib on PFS and OS was then investigated using subgroup analysis.

Statistical analysis was conducted using StataCorp 2019 Stata Statistical Software: Release 16.1 (College Station, Texas, USA), and R (The R foundation for Statistical Computing, version 4.1.0).

Results

The KNN Classifier Is Transferable to Randomized Trials Datasets

MONARCH 2 enrolled 669 women with endocrine resistant HR-positive HER2-negative ABC, 644 patients were eligible for KNN. At baseline, 411 patients (63.8%) were <65 years old, 473 (73.5%) had secondary endocrine resistance, 532 (82.7%) were post-menopausal. Bone-only disease was observed in 175 patients (27.2%), liver involvement in 173 patients (26.9%), and lung in 189 patients (29.4%) (Table 1).

Patients classified as pStage $IV_{aggressive}$ and pStage $IV_{indolent}$ were 183 (28%) and 461 (72%), respectively. All baseline features were equally balanced among pStage $IV_{aggressive}$ and pStage $IV_{indolent}$ apart from PR ($P < .001$), liver metastases ($P < .001$), and number of sites ($P < .001$) (Table 1).

Median PFS for pStage $IV_{aggressive}$ and pStage $IV_{indolent}$ was, respectively, 10.7 and 15.3 months ($P = .0011$) (Fig. 1A), while for OS, it was 47.8 and 32.2 months, respectively ($P < .0001$) (Fig. 1B).

Predicted CTCs Have an Independent Prognostic Impact in Terms of OS

After univariable analysis, the prognostic impact of predicted CTCs was observed for both PFS and OS (respectively

Table 1. Descriptive analysis of the 644 patients eligible for the KNN analysis. All baseline features were equally balanced among pStage $IV_{aggressive}$ and pStage $IV_{indolent}$ apart from progesterone receptor (PR) ($P < .001$), liver metastases ($P < .001$), and number of sites ($P < .001$).

	N	%	Indolent	Aggressive	*P*-value
Predicted CTCs			461 (71.58%)	183 (28.42%)	
Age					
<65	411	63.82	301	110	.217
≥65	233	36.18	160	73	
PR					
Negative	140	21.74	44	96	<.001
Positive	504	78.26	417	87	
Grade					
1	330	66.53	238	92	.368
2-3	166	33.47	126	40	
ECOG					
0	384	59.81	280	104	.330
1	258	40.19	179	79	
Endocrine resistance					
Secondary	473	73.45	344	129	.285
Primary	171	26.55	117	54	
Treatment arm					
Placebo	214	33.23	159	55	.281
Abemaciclib	430	66.77	302	128	
Menopausal status					
Post-menopausal	532	82.74	381	151	.923
Pre-menopausal	111	17.26	80	31	
Bone only					
No	469	72.83	326	143	.056
Yes	175	27.17	135	40	
Liver					
No	471	73.14	395	76	<.001
Yes	173	26.86	66	107	
Lung					
No	455	70.65	316	139	.063
Yes	189	29.35	145	44	
Number of sites					
1	257	39.91	219	38	<.001
2	192	29.81	136	56	
≥3	195	30.28	106	89	

HR = 1.39, 95% CI, 1.14-1.69, $P = .001$ and HR = 1.67, 95% CI, 1.33-2.10, $P < .001$), together with ECOG performance status (respectively HR = 1.43, 95% CI, 1.19-1.72, $P = .0001$ and HR = 1.68, 95% CI, 1.35-2.09, $P < .0001$), liver involvement (respectively HR = 1.62, 95% CI, 1.33-1.97, $P < .0001$ and HR = 1.74, 95% CI, 1.38-2.19, $P < .0001$) bone-only disease (respectively HR = 0.68, 95% CI, 0.55-0.84, $P = .0004$ and HR = 0.66, 95% CI, 0.51-0.85, $P = .0014$) and treatment arm (respectively HR = 0.55, 95% CI, 0.45-0.66, $P < .0001$ and HR = 0.76, 95% CI, 0.61-0.95, $P = .0177$) (Tables 2 and 3).

After multivariable Cox regression, predicted CTCs were confirmed as independently associated with prognosis in terms of OS (HR = 1.36, 95% CI, 1.03-1.79, $P = .0301$), together with ECOG performance status (HR = 1.74, 95% CI, 1.39-2.17, $P < .0001$), liver involvement (HR = 1.37, 95% CI, 1.03-1.82, $P = .0298$), and treatment arm (HR = 0.74, 95% CI, 0.59-0.93, $P = .01$) (Table 3).

Patients in the pStage $IV_{indolent}$ subgroup treated with abemaciclib experienced the best prognosis both in terms of PFS (Fig. 2A) and OS (Fig. 2B) (median PFS: 19.4 months, $P < .0001$; median OS: 55.5 months, $P < .0001$). On the other hand, patients in the pStage $IV_{aggressive}$ subgroup treated with placebo experienced the worst prognosis (median PFS: 7.1 months, $P < .0001$; median OS: 27.5 months, $P < .0001$) (Fig. 2A and 2B).

Predicted CTCs Can be Combined With Clinical Features to Explore Potential Subgroups Of Interest

The CTCs prediction was combined with clinically relevant and homogenous subgroups to highlight specific scenarios for hypothesis generation.

Among patients aged <65 years old, the pStage $IV_{indolent}$ subgroup experienced a significantly better OS compared to pStage $IV_{aggressive}$ (median OS 51.3 vs. 29.3 months, $P < .0001$) (Fig. 3A).

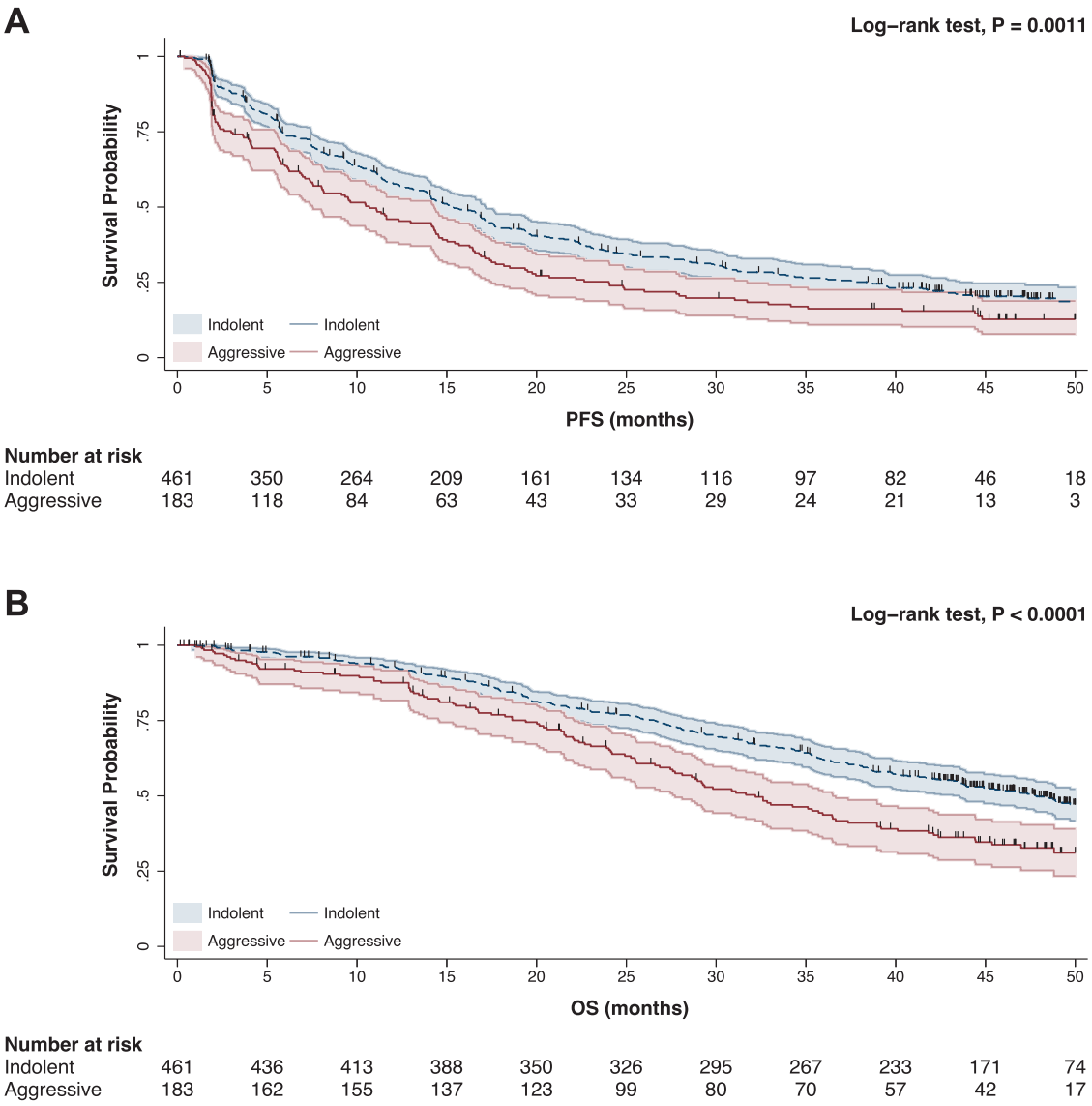


Figure 1. Kaplan Meier plot in terms of PFS (**A**) and OS (**B**) for CTCs prediction. Median PFS for pStage $IV_{aggressive}$ and pStage $IV_{indolent}$ was 10.7 and 15.3 months, respectively ($P = .0011$) (A) while for OS it was 32.2 and 47.8 months, respectively ($P < .0001$) (B).

The pStage $IV_{indolent}$ subgroup had a significantly favorable prognosis among patients with visceral (respectively median OS 44.9 vs. 28.8 months for pStage $IV_{indolent}$ and pStage $IV_{aggressive}$, $P < .0001$) (Fig. 3D) or liver metastases (respectively median OS 38.7 vs. 27.6 months for pStage $IV_{indolent}$ and pStage $IV_{aggressive}$, $P = .0263$) (Fig. 3E).

CTCs prediction was also able to significantly stratify patients regardless of previous treatment lines (no prior lines $P < .0001$; prior lines $P = .0299$) and endocrine sensitivity (primary resistance $P = .0112$; secondary resistance $P = .0004$) (Fig. 3B and 3C).

The treatment effect of abemaciclib on OS was then explored through subgroup analysis, showing a consistent benefit across all subgroups (Fig. 3G). In the pStage $IV_{indolent}$ subgroup, treatment had a HR = 0.68 (95% CI, 0.52-0.89), while a HR = 0.89 (95% CI, 0.60-1.35) was observed in the pStage $IV_{aggressive}$ subgroup, P for interaction was .262 (Fig. 3G).

Discussion

The present study investigated the potential of applying machine learning to a phase III clinical trial to predict the prognostic stratification of CTCs for hypothesis generation.

The previously published K-nearest neighbor (KNN) supervised machine learning algorithm was trained on a pooled dataset of 2436 ABC patient from EPAC and MDACC and applied to a real-world ABC cohort.[4,9] When applied to MONARCH 2, the model reproduced a comparable stratification. Patients in the pStage $IV_{aggressive}$ subgroup experienced an unfavorable prognosis both for PFS and OS, the latter confirmed after multivariable analysis.

Interestingly, the impact of abemaciclib was consistent in both subgroups, regardless of their baseline prognosis. On the other hand, CTCs prediction highlighted subgroups with a significantly different prognosis across clinically homogeneous features, such as visceral or liver involvement, where

Table 2. Uni- and multivariable prognostic analysis in terms of PFS.

	Univariable				Multivariable			
	HR	95% CI		*P*-value	HR	95% CI		*P*-value
ECOG PS								
0	1				1			
1	1.43	1.19	1.72	.0001	1.46	1.21	1.76	.0001
Tumor grade								
1	1							
2/3	1.22	0.98	1.51	.0702				
PR								
Negative	1							
Positive	0.90	0.72	1.13	.3671				
Liver involvement								
No	1				1			
Yes	1.62	1.33	1.97	<.0001	1.43	1.12	1.82	.0044
Bone-only involvement								
No	1				1			
Yes	0.68	0.55	0.84	.0004	0.7	0.52	0.94	.0176
Number of sites								
1	1				1			
2	1.15	0.92	1.43	.2292	0.87	0.66	1.15	.3351
≥3	1.48	1.19	1.84	.0004	0.92	0.68	1.23	.5618
Predicted CTCs								
Indolent	1				1			
Aggressive	1.39	1.14	1.69	.0011	1.23	0.98	1.56	.0793
Treatment arm								
Placebo	1				1			
Abemaciclib	0.55	0.45	0.66	<.0001	0.53	0.44	0.64	<.0001

pStage $IV_{indolent}$ experienced a comparable OS to patients without visceral or liver involvement.

This is in line with previous evidence, underlining the need for a more granular definition of MBC between indolent and aggressive, the latter subgroup being characterized not only by an unfavorable prognosis, but also by a specific mutational profile and organotropism.[12,13]

Notwithstanding the strong prognostic stratification achievable by CTCs enumeration, current clinical trials do not usually incorporate such characterization by design. An exploratory ancillary analysis of the TREnd trial analyzed the prognostic role of CTC enumeration and CTC RB1 gene expression in patients treated with palbociclib.[14] CTCs were analyzed by utilizing the CellSearch and DEPArray systems before treatment start, after the first cycle and at progression.[14] While the number of CTCs at baseline had no prognostic impact in terms of PFS or clinical benefit, patients with at least one detectable CTC at the first treatment cycle had a shorter PFS ($P = .02$).[14]

We previously demonstrated the role of CTCs in defining changes in biology, rather than treatment response, suggesting a more complex use scenario for CTCs.[15] Interestingly, TREnd showed how a CTCs increase between baseline and first cycle had an unfavorable impact on PFS ($P = .004$), and suggested a numerically favorable prognosis in patients with a detectable CTC RB1 expression at any timepoint, albeit this difference was not statistically significant.[14]

The potential of CTC-based biomarkers in HR-positive, HER2-negative ABC was also shown in a cohort of patients treated with everolimus and exemestane and characterized through methylation-specific (MS) qPCR applied to CTCs and, in a smaller subgroup (58 patients), matched plasma samples.[16] ESR1 methylation was found in 10 of 36 CTC-positive samples (27.8%), was highly consistent with the paired ctDNA samples, and was also linked to a lack of response to everolimus and exemestane.[16]

Combining CTC enumeration with additional genetic, epigenetic, and transcriptional characterizations is a promising approach in better highlighting different biological entities in ABC. Interestingly, although treatment benefit of abemaciclib was consistent across all subgroups, a numerically favorable OS was observed in pStage $IV_{indolent}$ patients with visceral involvement with respect to the overall population. Since pStage $IV_{indolent}$ and Stage $IV_{indolent}$ are usually characterized by a non-visceral organotropism, this subgroup may have been the result of specific targetable co-occurring alterations.[12,13,17] Moreover, an adequate prognostic and biological profiling may also enable personalized monitoring strategies, reducing both costs and patients' medicalization.[18,19]

Similarly, a ctDNA focused characterization of the MONARCH 2 study, explored the impact of abemaciclib in high-risk patients with a potentially more aggressive and resistant tumor biology defined through *PIK3CA* and *ESR1*

Table 3. Uni- and multivariable prognostic analysis in terms of OS. Predicted CTCs were confirmed as independently associated with prognosis in terms of OS together with ECOG performance status, liver involvement, bone-only disease, and treatment arm.

	Univariable				Multivariable			
	HR	95% CI		*P*-value	HR	95% CI		*P*-value
ECOG PS								
0	1				1			
1	1.68	1.35	2.09	<.0001	1.74	1.39	2.17	<.0001
Tumor grade								
1	1							
2/3	1.23	0.95	1.59	.1156				
PR								
Negative	1							
Positive	0.78	0.60	1.01	.0562				
Liver involvement								
No	1							
Yes	1.74	1.38	2.19	<.0001	1.37	1.03	1.82	.0298
Bone-only involvement								
No	1				1			
Yes	0.66	0.51	0.85	.0014	0.78	0.54	1.14	.205
Number of sites								
1	1				1			
2	1.31	0.99	1.72	.056	1.1	0.78	1.55	.5828
≥3	1.78	1.37	2.3	<.0001	1.24	0.87	1.78	.2305
Predicted CTCs								
Indolent	1				1			
Aggressive	1.67	1.33	2.10	<.0001	1.36	1.03	1.79	.0301
Treatment arm								
Placebo	1				1			
Abemaciclib	0.76	0.61	0.95	.0177	0.74	0.59	0.93	.01

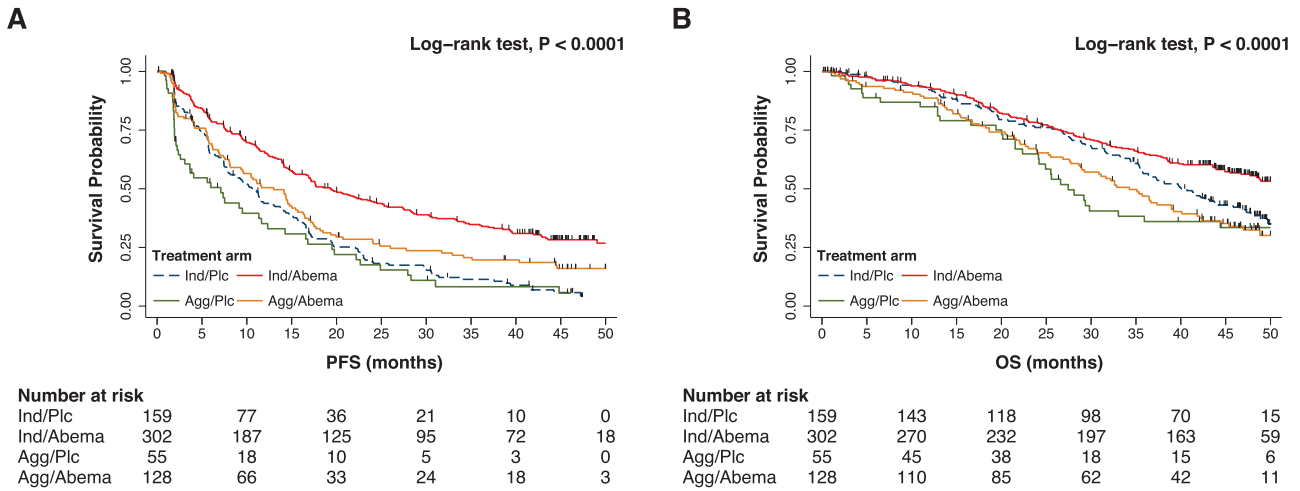
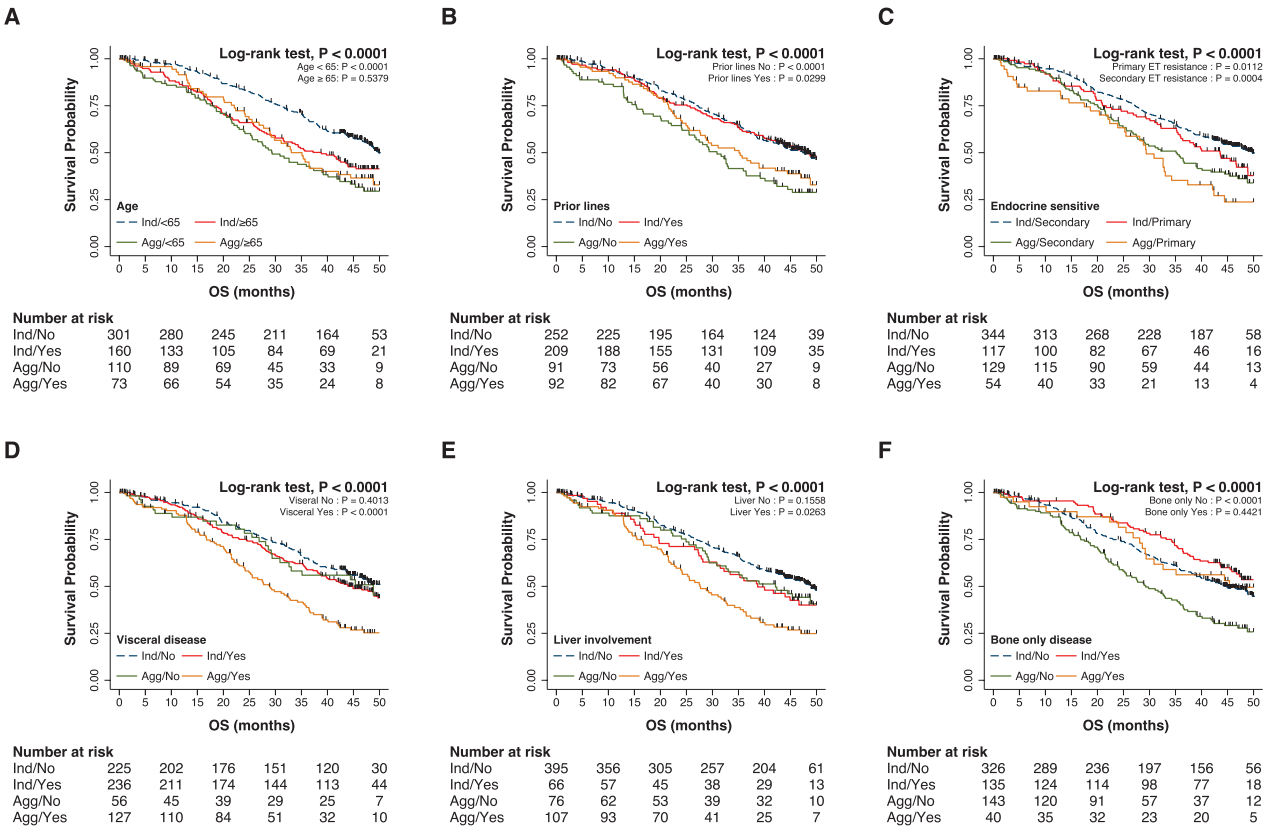


Figure 2. Kaplan Meier plot in terms of PFS (**A**) and OS (**B**) of CTCs prediction combined to treatment arm. pStage $IV_{indolent}$ (Ind) treated with abemaciclib (Abema) experienced the best prognosis both in terms of PFS (A) and OS (B) ($P < .0001$; $P < .0001$). Consistently, patients in the pStage $IV_{aggressive}$ (Agg) subgroup treated with placebo (Plc) experienced the worse prognosis.

mutational status.[20] Both the *PIK3CA*-wild-type and *PIK3CA*-mutant subgroups showed an improved PFS when compared to placebo plus fulvestrant (HR, 0.51; 95% CI, 0.33-0.78).[20] Consistently, also the *ESR1*-wild-type and *ESR1*-mutant subgroups showed improved PFS when compared to placebo plus fulvestrant, regardless of the presence of *PIK3CA* mutations.[20] Similar results were highlighted also for OS.[20] These results are in line with similar analyses performed with other



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Subgroup	N	HR (95% CI)	P for interaction
Modelled CTCs			
Indolent	461	0.68 (0.52, 0.89)	0.262
Aggressive	183	0.90 (0.60, 1.35)	
Endocrine resistance			
Secondary	473	0.79 (0.61, 1.03)	0.521
Primary	171	0.67 (0.44, 1.03)	
Number of sites			
1	257	0.83 (0.56, 1.23)	0.497
2	192	0.64 (0.43, 0.96)	
3+	195	0.88 (0.61, 1.27)	
Menopausal state			
Post-menopausal	532	0.76 (0.60, 0.97)	0.933
Pre-menopausal	111	0.74 (0.40, 1.36)	
Metastati sites			
Bone only	175	0.92 (0.57, 1.49)	0.479
Other	106	0.89 (0.50, 1.60)	
Visceral	363	0.68 (0.51, 0.90)	
Visceral involvement/CTC modelling			
Non visceral Aggressive	56	0.85 (0.37, 1.92)	0.207
Non visceral Indolent	225	0.91 (0.60, 1.39)	
Visceral Aggressive	127	0.87 (0.54, 1.40)	
Visceral Indolent	236	0.54 (0.37, 0.77)	
Overall	644	0.76 (0.61, 0.95)	

Figure 3. Kaplan Meier plot (**A**-**F**) and forest plot (**G**) in terms of OS for CTCs prediction and subgroups of clinical interest. In patients aged <65 years old (A), the pStage $IV_{indolent}$ (Ind) experienced a significantly better OS ($P < .0001$). CTCs prediction was able to stratify patients regardless of previous treatment lines (B) and endocrine sensitivity (C) (resistance $P = .0112$; secondary resistance $P = .0004$). Ind had a significantly favorable prognosis in patients with visceral (D) ($P < .0001$) or liver metastases (E) ($P = .0263$). Treatment effect of abemaciclib on OS was consistent across all subgroups (G).

CDK4/6i and suggest the intricate role of these drugs not only as synergistic ET partners but also as potential reversers of inherit or acquired ET resistance.[21] As CDK4/6i are making their way in the adjuvant setting, this aspects will be of crucial importance in shaping the future of the first- and second-line settings.[22]

Of note, the CellSearch system relies on an EPCAM-dependent enrichment which limits its characterization to epithelial cells, leaving out non-canonical cells such as epithelial to mesenchymal (EMT) CTCs.[23,24] EMT contributes to tumor heterogeneity and metastasis and is emerging as an independent predictive factor in MBC.[23,25] Being trained on a CellSearch-profiled cohort, the KNN model on one hand excludes any information deriving from the complete set of circulating cells subpopulations, but on the other inherits the strong and consolidated prognostic meaning of epithelial CTCs enumeration.

Our results, therefore, suggest how the prediction of canonical CTC enumeration can suggest possible scenarios for clinical trial optimization. Based on these findings, new biomarker-driven trials focused on the Stage $IV_{aggressive}$ subgroup may leverage the higher event probability on this population requiring a smaller sample size and a shorter follow-up.[26]

Conclusions

The study represents the first analysis of clinical outcome using predicted modeling of CTCs for Stage IV disease stratification. These hypothesis-generating results illustrate the need to expand resistance biomarkers evaluation in combination with CTCs stratification for improved biomarker-driven drug development and treatment personalization.

Funding

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Conflict of Interest

Lorenzo Gerratana reports consulting or advisory roles with AstraZeneca, Daiichi Sankyo, Eli Lilly, GlaxoSmithKline, Incyte, Novartis, Pfizer, Menarini Stemmline, and AbbVie; and research funding for Menarini Silicon Biosystems. Andrew Davis reports participating in a scientific advisory board for Pfizer, Inc. Fabio Puglisi Reports consulting or advisory roles with Roche, MSD, AstraZeneca, Novartis, Eli Lilly, Pfizer, Pierre Fabre, Daiichi Sankyo, Eisai, and Amgen, in addition to research funding from Eisai, AstraZeneca, and Roche. Massimo Cristofanilli reports consulting fees from Novartis, Menarini, Eli Lilly, Sermonix, G1 Therapeutics, Foundation Medicine, AstraZeneca, and Pfizer Inc., consulting/lecture fees from Foundation Medicine and Pfizer Inc., travel support from Foundation Medicine, participation on a Data Safety Monitoring Board or Advisory Board for Merck and AstraZeneca, and research funding from Pfizer Inc, Menarini, Eli Lilly, and G1 Therapuetics. Masha Kocherginsky reports royalties from a patent licensed by The University of Chicago to Corcept Therapeutics. The other authors indicated no financial relationships.

Author Contributions

Conception and design: L.G., M.K., M.C. Data acquisition: L.G., M.K., M.C. Quality control of data and algorithm: L.G., M.K. Interpretation of data, approval, and editing of the manuscript: All authors. All authors have read and approved the final version of the manuscript.

Data Availability

The data underlying this article were provided by Eli Lilly, Inc by permission. Data will be shared on request to the corresponding author with permission of Eli Lilly, Inc. Lilly's data sharing policies are provided on the clinicalstudydatarequest.com site under the Study Sponsors page.

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