Prediction of benefit from adjuvant pertuzumab by BluePrint RNA sequencing in the APHINITY trial

Ian E Krop[1], Lorenza Mittempergher[2], Joseph N Paulson[3], Fabrice Andre[4], Hervé Bonnefoi[5], Sherene Loi[6], Sibylle Loibl[7], Richard D Gelber[1], Carmela Caballero[8], Debora Fumagalli[8], Rajith Bhaskaran[2], Christa Dreezen[2], Rene Bernards[2], Laura J van 't Veer[2] and Martine J Piccart[9].

[1]Dana-Farber Cancer Institute, Boston, MA;[2]Agendia, Amsterdam, Netherlands; [3]Genentech, San Francisco, CA; [4]Gustave Roussy, Paris, France;[5]Bergonié Cancer Institute, Bordeaux, France; [6]Peter MacCallum Cancer Centre, Melbourne, Australia; [7]Goethe University, Frankfurt, Germany; [8]Breast International Group, Brussels

San Antonio Breast Cancer Symposium® - December 8-11, 2020, abstract #1672, PD3-01

Background

APHINITY is a phase III study (NCT01358877), which randomized 4805 patients with histologically centrally confirmed HER2+ early breast cancer (EBC) to standard adjuvant chemotherapy (C) and trastuzumab (T), plus either pertuzumab (P) or placebo[1] for one year. With a median follow up of 45 months, the primary analysis showed significant invasive disease-free survival (iDFS) benefit at 3 years with a hazard ratio (HR) of 0.81 (95% CI: 0.66-1.00; *P* = 0.045, 3-year IDFS percent 94.1% vs 93.2%) for the addition of P to C/T[1].

BluePrint (BP) is an 80-gene molecular subtyping test that classifies EBC into functional Basal, Luminal and HER2 BP-subtypes[3] according to gene expression. Previous studies showed that this genomic test can reclassify HER2+ EBC (as determined by IHC/FISH) into Basal, Luminal or HER2 BP-subtypes[4]. Recently, it was revealed that a smaller proportion of breast tumours may display two functionally activated BP pathways (BP dual activated subtypes, of which one is usually less pronounced), whereas the majority has clearly only one activated functional BP pathway (BP single activated subtypes)[5]. **We hypothesized that BluePrint could identify a subgroup of patients within the APHINITY population who derived further benefit from the addition of P to C/T.**

Methods

Genomic results were obtained using RNA sequencing (RNAseq) data[6] from a subset of APHINITY patients (N=969), which were derived from a 1023 unique patients nested case-control (NCC) set where event and matched controls were selected (1 iDFS event matched to 3 controls from the primary analysis database with 45 months median follow-up). Raw read counts were $\log_2$ transformed, followed by quantile normalization prior to genomic assessment. BP subtype scores were calculated in the same manner to the standard microarray diagnostic testing and calibrated based on a bridge analysis with matched microarray and RNAseq data. IDFS outcome based on genomic subtype and the treatment arm (P+C/T *vs* placebo + C/T) was analyzed. Results are reported descriptively with 95% CIs.

Results

Table 1

BP	Total	Treatment Arm		Nodal Status		Age		Anthracycline containing regimen		IDFS Range (months)
		Trastuzumab +placebo	Trastuzumab + Pertuzumab	Negative	Positive	<35yr	≥35yr	No	Yes	
Basal	210	112 (53.3%)	98 (46.7%)	40 (19.0%)	170 (81.0%)	8 (3.8%)	202 (96.2%)	43 (20.5%)	167 (79.5%)	(0.36, 56.61)
Luminal	484	247 (51.0%)	237 (49.0%)	90 (18.6%)	394 (81.4%)	35 (7.2%)	449 (92.8%)	86 (17.8%)	398 (82.2%)	(0.72, 58.02)
HER2	275	156 (56.7%)	119 (43.3%)	39 (14.2%)	236 (85.8%)	22 (8.0%)	253 (92.0%)	43 (15.6%)	232 (84.4%)	(1.97, 57.17)
Total	969	515	454	169	800	65	904	172	797	

Figure 1

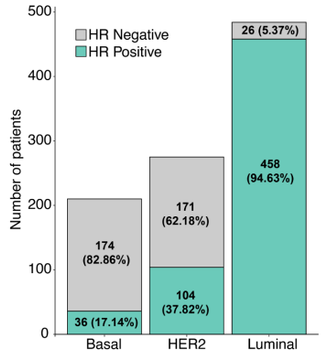
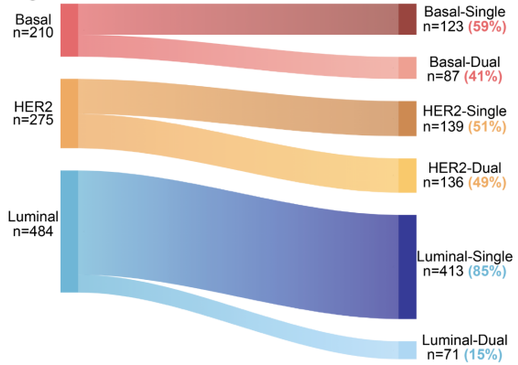


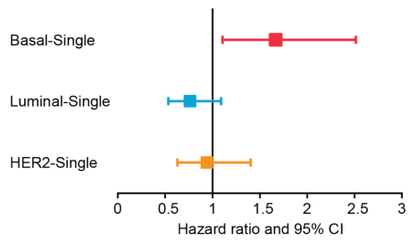
Figure 2



From the 969 patients within the NCC subset, BP subtyping classified 210 (22%) as Basal, 484 (50%) as Luminal, and 275 (28%) as HER2 (**Table 1**). A majority of patients in all BP groups were lymph-node positive, over 35 years old, and received an anthracycline containing regimen (**Table 1**). Most patients in the BP Luminal group were HR positive whereas a majority of BP Basal and BP HER2 patients were HR negative (**Figure 1**).

Figure 3

A. Prognostic analysis



B. Predictive analysis comparing effect of **P+T vs T**$_{+placebo}$

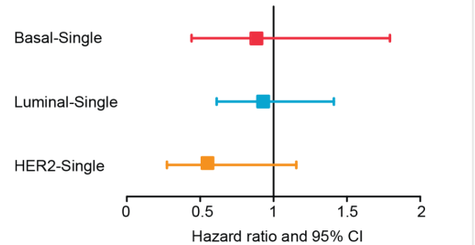


Table 2

Multivariate Model		BP – Type (single vs other)	Treatment Arm (P+T vs T$_{+placebo}$)	HR Status (pos vs neg)	Nodal Status (pos vs neg)	Age (≥35yr vs <35yr)	Anthracycline (yes vs no)
Basal	Hazard Ratio	**1.67**	0.84	1.02	**4.53**	0.85	0.87
	95% CI	**1.11 – 2.51**	0.64 – 1.09	0.75 – 1.38	**3.10 – 6.62**	0.51 – 1.42	0.61 – 1.24
Luminal	Hazard Ratio	0.76	0.83	1.06	**4.51**	0.88	0.86
	95% CI	0.53 – 1.09	0.64 – 1.08	0.74 – 1.52	**3.09 – 6.58**	0.52 – 1.47	0.61 – 1.23
HER2	Hazard Ratio	0.94	0.83	0.88	**4.49**	0.85	0.87
	95% CI	0.63 – 1.40	0.64 – 1.09	0.66 – 1.17	**3.08 – 6.56**	0.51 – 1.43	0.61 – 1.23

Further dissection of BP results revealed that a large majority of Luminal and a majority of Basal subtypes were single-activated. In contrast, about 50% of HER2 subtypes were single-activated (**Figure 2**). After NCC-inverse probability weighted corrected multivariate Cox regression analysis, no significant differences in iDFS were observed among the different genomic subtypes (dual and single activated subtypes included). Multivariate survival analysis among the BP single molecular subtypes indicated that patients with a BP Basal single-activated tumor are significantly more likely to have an event compared to other subtypes (**Figure 3A, Table 2**). Of note, among other variables included in the multivariate model, only nodal status appeared to be associated with IDFS (**Table 2**).

Importantly, based on predictive analysis, there was a trend of greater benefit with the addition of pertuzumab to trastuzumab in patients with BP HER2 single-activated tumors compared with other groups (**Figure 3B**).

Conclusion

In this exploratory analysis, HER2+ tumors with a BluePrint single transcriptional HER2 activated pathway showed a trend for greater benefit from pertuzumab than tumors in which multiple mitogenic pathways, or in which single luminal or basal pathway are activated. Notably, in this pathologically confirmed HER2+ set, BluePrint single-activated Basal-type patients appeared to show a significant worse prognosis compared to other single-activated types. Further research is ongoing to confirm these findings and to further define the role of BluePrint in the characterization of HER2 + tumors.

References

1. Von Minckwitz G. et al, NEMJ, 2017
2. Piccart M. et al, SABCS, 2019
3. Krijgsman O. et al, BCRT, 2011
4. Whitworth P. et al, ASCO, 2014
5. Kuilman M. et al, EBCC12, 2020
6. Krop IE et al, ASCO, 2020

This presentation is the intellectual property of Ian Krop and Lorenza Mittempergher. Contact them at Ian_Krop@dfci.harvard.edu and lorenza.mittempergher@agendia.com for permission to reprint and/or distribute.