Adding precision to 2018 ASCO/CAP HER2 testing guidelines in breast cancer with genomic profiling

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BACKGROUND

- Biological heterogeneity of HER2 positive breast cancers is supported by a modest benefit of HER2-targeted therapies reported in the APHINITY and ExteNET trials. This highlights the need for improved biomarkers that more precisely identify patients who benefit from HER2-directed agents.
- The 80-gene molecular subtyping signature, BluePrint (BP), classifies breast tumors into Luminal, HER2 or Basal subtype based on the gene expression of downstream signaling pathways (1). Previous work showed a substantial proportion of tumors identified as HER2 equivocal or HER2 positive by 2013 ASCO/CAP guidelines may be reclassified as non-HER2 subtype by BP (2).
- In 2018, ASCO/CAP HER2 IHC/ISH classification guidelines were revised to reduce the frequency of HER2 equivocal cases, for which treatment recommendations have been ambiguous (3). Here we evaluated concordance between HER2 status by 2018 ASCO/CAP guideline classification and BP molecular subtyping.

METHODS

- Pathology reports are provided by physicians for samples that are tested with the 70-gene risk of distant recurrence signature, MammaPrint (MP), and BluePrint as part of routine diagnostic care.
- BP indices for each subtype (Luminal, HER2, and Basal) were calculated based on expression of 80 genes and range from -1.6 to +1.6. The subtype with the highest index of the three is the dominant molecular subtype reported for the tumor. MP was used to further stratify BP Luminal into Luminal A and Luminal B.
- This analysis includes breast tumor specimens sent to Agendia (Irvine, CA) from January 2019 to January 2020. HER2 IHC/ISH results, which were determined locally based on ASCO/CAP 2018 guidelines, were available for 1453 samples. Specimens were obtained by either core biopsy (n=656), surgical excision (n=796), or information was unavailable (n=1). There was no significant difference in MP stratification between samples obtained by these methods.

RESULTS

- **Comparison between HER2 IHC/ISH and BP subtyping.** Of 1453 samples, 1336 (92%) were HER2 negative, 99 (7%) were HER2 positive, and 18 (1.2%) were HER2 equivocal under 2018 guidelines (**Table 1**). BP reclassified 17 of 99 (17%) HER2 positive tumors as Luminal A, 40 of 99 (40%) as Luminal B, and 11 of 99 (11%) as Basal; the remaining 31% were confirmed HER2 (**Table 1 and Figure 1**). Furthermore, 55 of 99 (55%) HER2 positive tumors were also ER and PR+ by IHC, with 48 (87%) of these reclassified as BP Luminal. Of HER2 equivocal tumors, 16 of 18 (89%) reclassified as Luminal and 2 of 18 (11%) as Basal. Of HER2 negative tumors, BP classified 96 of 1336 (7%) as Basal and 2 of 1336 (0.15%) as HER2.
- **Evaluation of BP indices in tumors classified as HER2 by IHC/ISH.** BP indices for each subtype were calculated for HER2 positive tumors that were reclassified to Luminal by BP (**Figure 2A**), classified as HER2 by BP (**Figure 2B**), and reclassified as Basal by BP (**Figure 2C**). The dominant molecular subtype is reported based on the highest BP index. Of HER2 positive tumors that reclassified as BP Luminal, 13 of 56 (1 patient excluded due to unknown HR status) had positive indices for both BP HER2 and BP Luminal (**Figure 3**).

BluePrint reclassified 69% of HER2 positive tumors to non-HER2 molecular subtypes, suggesting these tumors may have suboptimal responses to HER2-directed therapy compared to HER2 enriched.

Molecular classification by BluePrint adds further precision in classifying HER2 positive patients and potential to predict responsiveness to HER2-targeted therapies.

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REFERENCES
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TABLES and FIGURES

Table 1. IHC/ISH and BP classification

IHC/ISH Status	BP Luminal A	BP Luminal B	BP HER2	BP Basal	Total
HER2 Positive	**17**	**40**	**31**	**11**	**99**
IHC ER+PR+	15	33	7	0	
IHC ER+ or PR+	2	6	13	2	
IHC HR-	0	0	11	9	
HR unknown	0	1	0	0	
HER2 Equivocal	**9**	**7**	**0**	**2**	**18**
IHC ER+PR+	9	7	0	0	
IHC HR-	0		0	2	
HER2 Negative	**681**	**557**	**2**	**96**	**1336**
IHC ER+PR+	619	470	1	12	
IHC ER+ or PR+	50	75	1	27	
IHC HR-	6	4	0	55	
HR unknown	6	8	0	2	

Figure 1. Reclassification of HER2 IHC/ISH by BP

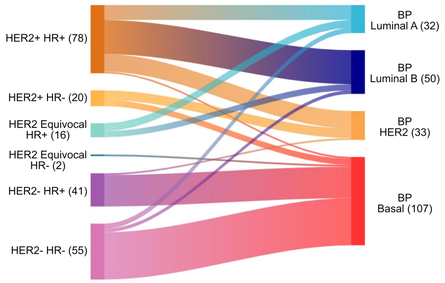
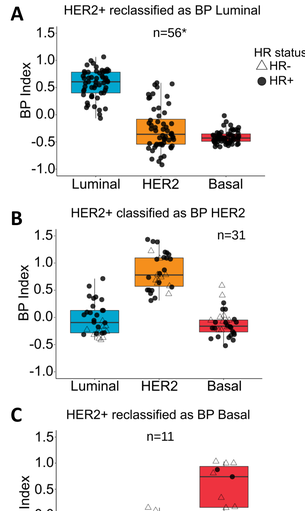


Figure 1 excludes patients with unreported MP result, unknown HR status, or HER2 negative patients who are BP Luminal (n=1241).

Figure 2. Evaluation of BP indices in tumors classified as HER2+ by IHC/FISH



HR status in Figure 2 includes tumors that were ER and/or PR+.
* One patient excluded due to unknown HR status.

Figure 3. Dual positivity of HER2 and Luminal BP indices in tumors classified as HER2+ by IHC/ISH.