HER2 Discordant Results in Local vs. Central Testing in the Phase 3 Nelipepimut-S Trial and Implementation of Leica Bond Oracle HER2 Immunohistochemistry (IHC) System for Low and Intermediate Levels (1+, 2+) of HER2 Protein Expression as a Companion Diagnostic

M Melisko MD[1], E Mittendorf MD PhD[2], S Safina MD PhD[3], M Schenker MD PhD[4], AM Brunt MD[5], M Litwiniuk MD[6], J Mackey MD[7], K Petrakova MD[8], S Alieva MD[9], L Chance[10], GS Choy PharmD[10], L Morgenthien PhD[11], S Kumar PhD[12], HS Rugo MD[1]

[1]University of California, San Francisco, San Francisco, CA, USA; [2]University of Texas, M.D. Anderson Cancer Center, Houston TX, USA; [3]Republican Clinical Oncology Center, Kazan, Republic of Tatarstan, Russian Federation; [4]Sf Nectarie Oncology Center, Craiova, Romania; [5]University Hospital of North Staffordshire, Stoke on Trent, UK; [6]Greater Poland Cancer Center, Poznan, Poland, [7]Cross Cancer Institute, Edmonton, Alberta Canada; [8]Masaryk Memorial Cancer Institute, Cancer Care Clinic, Brno, Czech Republic; [9]Donetsk Regional Antitumor Center, Donetsk, Ukraine; [10]Galena Biopharma, Inc., Portland OR, USA; [11]ICON, NC, USA; [12]Leica Biosystems, Danvers Massachusetts, USA.

Background

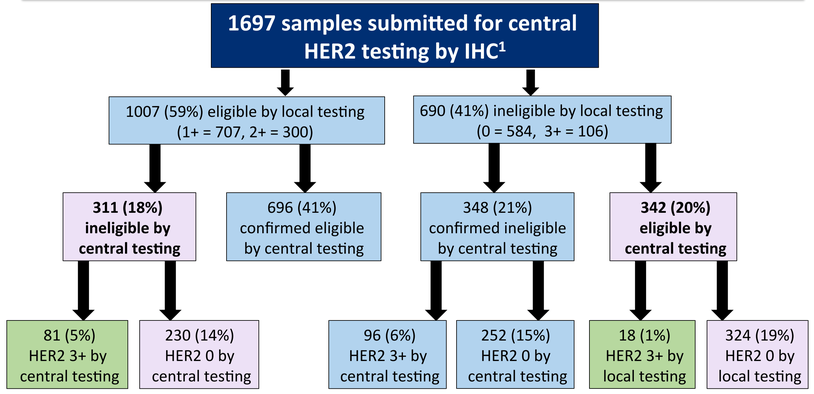
- Significant discordance exists between local and central laboratory testing for HER2 overexpression
- Discordance rates for tumors selected for HER2 0-2+ are not well described
- It is important to document sources and rates of discordance to avoid under and over treatment with HER2-targeted therapies

PRESENT Study

- NeuVax™ is a peptide vaccine that targets cells expressing any level of HER2
- The PH3-01, **PRESENT** (**P**revention of **R**ecurrence in **E**arly **S**tage, Node Positive Breast Cancer with Low to Intermediate HER2 **E**xpression with **NeuVax™ T**reatment) study is an international, multi-center randomized double-blind trial of NeuVax™ in 700 patients (NCT01479244)
- Patients are randomized to receive NeuVax™ + adjuvant vs. placebo + adjuvant over 36 months
- Study is open for enrollment in the US, Canada, UK, Germany, France, Israel, Russia, Ukraine, Poland, Romania, Hungary, Czech Republic, and Bulgaria
- **Objectives and Endpoints:**
  - Assess efficacy/safety of vaccine NeuVax™
  - Endpoints: DFS, OS, time to recurrence
- **Eligibility:**
  - Early-stage, node-positive breast cancer
  - Completed adjuvant chemotherapy within 12 wks
  - HLA-A2 or A3 positive
  - Low/intermediate expression of HER2 protein (1+ or 2+ by IHC; FISH non-amplified)
- **Methods: Screening for HER2 Expression:**
  - Local HER2 testing: samples for any degree of HER2 by local results to be assessed by central laboratory for confirmation of eligibility
  - Initial HER2 central testing: central laboratory confirmation using DAKO HercepTest™
  - Current HER2 central testing: upon approval of protocol amendment, patients are evaluated with Leica BOND Oracle III for HER2 eligibility

Results

Figure 1. Comparison of Local vs. Central IHC (N = 1697)*



= No discordance or discordance did not affect eligibility

= Discordance resulted in change in eligibility

= Change in eligibility was to/from HER2 3+

[1]**Eligibility definitions**: Local testing: Any degree of HER2 testing by IHC is eligible for screening

Central testing: HER2 1+ or 2+ by DAKO HercepTest™ are eligible for enrollment

- Overall discordance rate = 54% (921/1697)
- Discordance resulted in changes in eligibility for 38% of patients
  - 311 patients (18%) were not enrolled due to discordance between local and central results

Table 1. Global IHC Discordance Rates (N = 1697)*

Local Testing	Central Testing (DAKO HercepTest™) 0 (n = 482)	1+ (n = 731)	2+ (n = 307)	3+ (n = 177)	Discordance Rate
0 (n = 584)	245 (42%)	247 (42%)	77 (13%)	15 (3%)	**58%**
1+ (n = 707)	182 (26%)	362 (51%)	132 (19%)	31 (4%)	**49%**
2+ (n = 300)	48 (16%)	114 (38%)	88 (29%)	50 (17%)	**71%**
3+ (n = 106)	7 (7%)	8 (8%)	10 (9%)	81 (76%)	**24%**
Discordance Rate	49%	50%	71%	54%	

*Data cut-off: October 14, 2014

Table 2. IHC Discordance Rates by Location

Region	Country	Discordance Rate (n/N)	
North America	US	46% (86/185)	47% (102/219)
	Canada	47% (16/34)	
Eastern Europe	Bulgaria	70% (62/89)	55% (726/1309)
	Czech Republic	50% (55/109)	
	Hungary	58% (48/83)	
	Poland	60% (25/42)	
	Romania	59% (48/82)	
	Russia	53% (287/539)	
	Ukraine	55% (201/365)	
Western Europe	France	38% (5/13)	55% (79/143)
	Germany	60% (28/47)	
	UK	55% (46/83)	
Middle East	Israel	54% (14/26)	54% (14/26)

- Discordance (local vs. central) rates varied by geographic location (Table 2)
  - Ranged from 38% to 70% by country
  - Discordant rates were comparable and similar across regions
  - Rates of patients who were locally ineligible but proved to be eligible by central lab ranged from 29% (Israel) to 67% (UK, Canada)

LEICA Bond Oracle HER2 IHC Assay

- The Leica Bond Oracle HER2 IHC System is an FDA-approved fully-automated assay to accurately determine HER2 oncoprotein status in breast cancer tissue as an aid in the assessment of patients for whom Herceptin® treatment is being considered
- The Leica assay include four cell line controls, each one with a unique staining pattern representing the four HER2 classification (0, 1+, 2+, 3+)
- The Leica Bond Oracle HER2 IHC assay is also validated to reliably identify IHC HER2 1+ and 2+ cases in order to support the on-going PRESENT trial as a companion diagnostic for NeuVax™; whereas, the DAKO HercepTest is not validated to distinguish between HER2 IHC 0 and HER2 1+

Table 3. IHC DAKO vs. LEICA**

		Leica Bond Oracle 0 n = 26	1+ n = 138	2+ n = 24	3+ n = 1
DAKO HercepTest™	0 n = 16	6 (23%)	10 (7%)	0 (0%)	0 (0%)
	1+ n = 117	17 (65%)	91 (66%)	9 (38%)	0 (0%)
	2+ n = 56	3 (12%)	37 (27%)	15 (63%)	1 (100%)
	3+ n = 0	0 (0%)	0 (0%)	0 (0%)	0 (0%)

** Data cut-off: October 29, 2014

Conclusions

- Significant discordance exists between local and central laboratory test results for HER2 expression by IHC, even at lower levels of expression
- Discordance is similar across geographic regions
- Current available tests (DAKO HercepTest™) are defined by their ability to determine HER2 3+ status by IHC
- To improve accuracy and specificity for HER2 1+ and 2+ status testing and investigate a companion diagnostic for NeuVax™, the Leica Bond Oracle HER2 IHC System has been incorporated as central HER2 IHC screening for the PRESENT study
- Preliminary early LEICA results demonstrate high to moderate one-way concordance in HER2 1+/2+ compared to DAKO HercepTest™

The authors would like to thank the investigators and patients participating in the PRESENT study

The PRESENT study is sponsored by Galena Biopharma, Inc.

Disclosures:
HS Rugo serves on the Scientific Advisory Board for Galena Biopharma, Inc.
All other investigators receive research funding from Galena Biopharma, Inc. for the conduct of the PRESENT study.
L Chance and G Choy are employed by Galena Biopharma, Inc.; L Morgenthien is employed by ICON.

GALENA BIOPHARMA