Diverse Inter- and Intra-patient Circulating Tumor Cells (CTCs) Phenotypic Heterogeneity Identified Across Multiple Metastatic Breast Cancer (mBCa) Cohorts
Beverly Horn, Adam Jendrisak, Joseph Schonhoff, Ryan P. Graf, Priscilla Ontiveros, Megan Kearney, Yipeng Wang, Mark Landers, Ryan Dittamore
Epic Sciences, 9381 Judicial Dr, Suite 200, San Diego, CA 92121 ● epicsciences.com

Background
• There is an unmet need for biomarkers to guide 1x selections for mBCa patients among choice of hormone therapies, targeted therapies and chemotherapy.
• We previously developed a quantitative measure of CTC phenotypic heterogeneity in mCRPC, and found that high heterogeneity patients are associated with relatively better overall survival on chemotherapy, while low heterogeneity patients have better survival with AR signaling inhibitors (Schen et al. 2017 Cancer Research).
• Here, the same methodology was applied to mBCa patient cohorts to ascertain the feasibility of CTC heterogeneity analysis in mBCa.

Methods
• 198 blood samples from mBCa patients were processed for CTC analysis utilizing the Epic Sciences platform. Following enumeration, multi-dimensional phenotypic characterization analysis was performed utilizing protein expression and digital pathology analysis.
• Features from each CTC (5158 CTCs from 198 patients, 107+HR+, 14 HER2+, 88+HER2+, 69 TNBC) were clustered using unsupervised approach (K-means) and the optimal number of clusters was determined using the elbow method with greater than 85% variance taken into account. Shannon index was used to assess the intrapatient heterogeneity. 1ST CTCs from various classified CTC subtypes were single cell sequenced for copy number variations.

Morphological Features Associated with CTC Subtypes

Example Cell Images of CTC Subtypes

High CTC Phenotypic Heterogeneity is Common in mBCa Blood Samples

Phenotypic CTC Subtypes are Associated with Distinct Genomic Alterations

CTC Subtypes are Associated with Breast Cancer Pathological Types

Conclusions
• Diverse inter- and intrapatient phenotypic CTC heterogeneity is observed across multiple cohorts with specific genome profiles detected for different CTC subtypes.
• We seek to determine if patients with low heterogeneity might be better candidates for hormonal and targeted therapy.
• Studies linking heterogeneity to therapeutic efficacy and patient outcome are ongoing.