Estrogen Receptor mRNA Levels in Breast Cancer Predicts Response to Tamoxifen
Jennifer M. Bordeaux¹, Huan Cheng¹, Allison W. Welch¹, Bruce G. Haffty¹, Donald R. Lannin¹, Xingyong Wu², Nan Su², Xiao-Jun Ma², Yuling Luo² and David L. Rimm¹
¹Department of Pathology, Yale School of Medicine, New Haven, CT 06520, USA
²Advanced Cell Diagnostics, Hayward, CA 94545, USA

ABSTRACT

Two tissue microarray (TMA) cohorts of archival breast cancer samples from Yale were used in this study. The Yale Sentinel Node Cohort, called YMTA 128 (patients diagnosed from 2002-2006, n = 238) and an independent and non-overlapping cohort, called YMTA 130, from patients diagnosed from 2006-2005 (n = 524). Clinicopathologic characteristics of both cohorts are found in Table 1.

RESULTS

• The RNAscope assay combined with AQUA quantification specifically and reproducibly measures ESR1 in FFPE breast cancer tissues.
• ER mRNA and ER protein measured by AQUA have a non-linear relationship.
• High ESR1 predicts tamoxifen response whereas low ESR1 does not.
• ESR1 levels were not prognostic at any cutpoint.

MATERIALS & METHODS

METHODS: Messenger RNA for ER (ESR1) and Ubiquitin C (Ubc) were visualized using RNAscope probes and levels were quantified by quantitative in situ hybridization (qISH) on two Yale breast cancer cohorts on tissue microarrays. ESR1 levels were compared to ER protein levels measured by QIF using the SP1 antibody.

RESULTS: ESR1 mRNA is reproducibly and specifically measurable by qISH on tissue collected from 1993 or later. ESR1 levels were correlated to ER protein levels in a non-linear manner on two Yale cohorts. High levels of ESR1 were found to be predictive of response to tamoxifen.

CONCLUSION: Quantification of mRNA using qISH may allow assessment of large cohorts with minimal formalin fixed, paraffin embedded tissue. Exploratory data using this method suggests that measurement of ESR1 mRNA levels may be predictive of response to endocrine therapy in a manner that is different from the predictive value of ER. Further studies are underway using tissue microarrays from other institutions to determine how tissue age affects this assay.

BACKGROUND

Despite the usefulness of estrogen receptor (ER) as a predictive marker for endocrine therapy 50% of ER positive patients still recur, indicating a need for additional predictive biomarkers for endocrine therapy [1]. Assessment of mRNA expression signatures allows for the comparison of thousands of genes at a time. As a result, mRNA expression-based signatures have shown that better patient stratification can be achieved by looking at many genes [2,3]. However, Paik and colleagues have suggested that even looking at the mRNA from a single gene could show predictive power [4]. Recently, a novel mRNA in situ hybridization (ISH) technique called RNA-scope (Advanced Cell Diagnostics, Inc.) has been developed that can be used to detect RNA transcripts on formalin-fixed paraffin embedded (FFPE) tissue [5]. Here we modified the AQUA method for quantitative measurement of protein to combine it with the RNAscope method to quantify ER mRNA in situ and to compare to ER protein levels determined by quantitative immunofluorescence (QIF) on two breast cancer cohorts.

REFERENCES