Microarray analysis of vascular endothelial growth factor (VEGF)-dependent angiogenic biomarkers in squamous cell carcinoma (SCCA) and adenocarcinoma (AC) of the cervix

Rebecca Feldman, Ph.D.1, Zoran Gatalica MD, DSc1, Sandeep Reddy MD1, Krishnansu Tewari, M.D., F.A.C.O.G., F.A.C.S.2
1Caris Life Sciences, Phoenix, AZ; 2University of California, Irvine Medical Center, Orange, CA

Abstract (#3886)

Objective: On August 14, 2014, the US Food & Drug Administration approved bevacizumab for advanced cervical cancer based on Gynecologic Oncology Group (GOG) protocol 240, the randomized phase-3 clinical trial which demonstrated significantly improved overall survival with combination chemotherapy plus bevacizumab compared to chemotherapy alone. Although the signal for efficacy of bevacizumab was not observed in a subgroup analysis of prognostic factors for ACs, this histotype is comprised only 20% of the GOG 240 population. We sought to determine whether VEGF pathway biomarkers were differentially expressed between SCCA and AC of the cervix.

Methods: 244 cervical cancer cases referred to Caris were examined retrospectively for changes in gene expression by the Agilent microarray platform. Among the biomarkers studied were VEGFA ligand, VEGF receptors (VEGFR1, VEGFR2), the positive regulator of the VEGF-dependent angiogenic pathway, hypoxia-inducible factor 1 alpha (HIF1α), and the negative regulator von Hippel-Lindau or VHL (involved in ubiquitination and degradation of HIF1α). The 2-tail Fisher’s exact test was performed to test where proportions of positive results were different by subgroup (p<0.05). ANNOI D (IAS Institute Inc., Cary, NC) was utilized for statistical analysis.

Results: The median age for the 158 (65%) cases of SCCA was 47 yrs and that of the 86 (35%) ACs was 45 yrs. Overexpression of VEGFR1/2 was not observed in either cell type (0-2%). Importantly, overexpression of the VEGF ligand was found in 72% and 68% of SCCA and AC, respectively (p<0.001). HIF1α was overexpressed in 52% of SCCAs and 43% of ACs (p<0.05). VHL was overexpressed in only 26% of SCCAs and 30% of ACs. Co-expression of HIF1α and VHL was present in 75% of both SCCA and AC cases.

Conclusions: These data suggest that biomarkers along the VEGF-dependent path of tumor angiogenesis are not differentially expressed between SCCA and AC of the cervix. LIGand binding and sequestration using the monoclonal antibody, bevacizumab, is likely to inhibit angiogenesis in women suffering from advanced SCCA or AC of the cervix. The extent to which pre-treatment microarray analysis to guide therapy decisions requires further investigation.

Background

On August 14, 2014, Bevacizumab, the first biological/targeted therapy, was approved for cervical cancers; a disease with limited therapy options after progression on platinum-based regimens. We examined the VEGF pathway for differential expression between SCCA and AC of the cervix.

Methods

Two-hundred forty four cases referred to Caris Life Sciences from 2000 through 2012 were examined retrospectively for changes in gene expression by the Agilent microarray platform. Among the biomarkers studied were VEGFA ligand, VEGF receptors (VEGFR1, VEGFR2), the positive regulator of the VEGF-dependent angiogenic pathway, hypoxia-inducible factor 1 alpha (HIF1α), and the negative regulator von Hippel-Lindau or VHL (involved in ubiquitination and degradation of HIF1α). The 2-tail Fisher’s exact test was performed to test where proportions of positive results were different by subgroup (p<0.05). ANNOI D (IAS Institute Inc., Cary, NC) was utilized for statistical analysis. A histologic analysis of predictive biomarkers by immunohistochemistry (Caris Life Sciences) was performed for additional insight into potential bevacizumab combination strategies.

Results

Figure 1a. Histology (n=244)

Figure 1b. Disease Status (n=244)

Figure 2. VEGF-dependent angiogenic pathway

Figure 3a. Expression across subtypes (AC-SCCA)

Figure 3b. Histology: AC (green) vs. SCCA (red)

Figure 3c. Disease Status: Primary  (blue) vs. Metastatic (green)

Figure 4. mRNA expression in VEGFA + vs. VEGFA – patients.

Table 2. Genes along Angiogenic Pathway, including oncogenic recetpor tyrosine kinases

Table 1. Sites of Metastasis

Table 2 and Figure 4. Oncogenes and Angiogenesis. VEGF expression is upregulated through activation of tyrosine kinase receptors including EGFR, HER2, PDGFR and through activation of the AKT and MAPK pathways. mRNA expression patterns of these genes and biomarkers involved in angiogenesis were assessed for differences between VEGFA+ and VEGFA– cervical cancer patients. VEGF+ status was statistically correlated with overexpression of c-MET (75% vs. 52%; p=0.044) and PDGFC (16% vs. 1%; p=0.001). ERBB2 and PTEN isoforms (ECOD) overexpression correlation with VEGFA+ status was trending towards statistical significance as well.

Conclusions

These data suggest that biomarkers along the VEGF-dependent pathway of tumor angiogenesis are not differentially expressed between SCCA and AC of the cervix.

Overexpression of the ligand, VEGF, and the receptor, VEGFR1/2 modulates angiogenesis in cervical cancer.

Ligand binding and sequestration using the monoclonal antibody, bevacizumab, is likely to inhibit angiogenesis in women suffering from advanced SCCA or AC of the cervix.

The extent to which pre-treatment microarray analysis to guide therapy decisions requires further investigation.

References

Contact email: rfeldman@carisls.com