FGF10 is differentially expressed in high-grade serous ovarian cancers.

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Ovarian cancer is the most lethal gynecologic cancer¹. Soluble factors are an important part of the tumor microenvironment². We sought to identify genes associated with high-grade serous ovarian cancer (HGSC) by comparing global gene expression profiles of normal ovary with that of primary tumors from women diagnosed with HGSC using published microarray data^{3,4}. We found significant differential expression of the morphogen fibroblast growth factor 10 (FGF10) in high-grade serous ovarian tumors.

Keywords: ovarian cancer, high-grade serous ovarian cancer, HGSC, targeted therapeutics in ovarian cancer, systems biology of ovarian cancer, FGF10.

1 The five-year survival rate for women diagnosed with high-grade serous ovarian cancer
2 is between 30-40% and has not changed significantly in decades^{8,9}. The development of novel,
3 targeted therapeutics to treat HGSC can be facilitated by an enhanced understanding of the
4 transcriptional behavior of ovarian tumors relative to that of the normal ovary. We mined
5 published microarray data^{3,4} to compare global gene expression profiles between HGSC ovarian
6 tumors and that of normal ovarian tissue. We identified the gene encoding fibroblast growth
7 factor 10, FGF10, as among the most differentially expressed in HGSC tumors of the ovary.
8 FGF10 may be a gene of interest when prioritizing the study of target genes and pathways for
9 the development of novel therapeutic interventions in high-grade serous ovarian cancers.
10

11 12 **Methods**

13 We used microarray data from datasets GSE124766³ and GSE146556⁴ for this
14 differential gene expression analysis of high-grade serous carcinomas. The Benjamini and
15 Hochberg method of p-value adjustment was used for ranking of differential expression but raw
16 p-values were used for assessment of statistical significance of global differential expression.
17 Log-transformation of data was auto-detected, and the NCBI generated category of platform
18 annotation was used. GSE146556⁴ was generated using Affymetrix Human Gene 1.0 ST Array
19 with n=3 for normal ovarian tissue and n=40 for tumors from patients with high-grade serous
20 ovarian cancer. GSE124766³ was generated using Agilent-014850 Whole Human Genome
21 Microarray 4x44K G4112F with n=3 of for normal ovarian tissue and n=8 for tumors from
22 patients with high-grade serous ovarian cancer. GEO2R provides mRNA expression levels only
23 for the top 250 most differentially expressed genes.
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25 A statistical test was performed to evaluate whether FGF10 expression was significantly
26 different when comparing normal ovarian tissue from control subjects and primary tumors from
27 women diagnosed with HGSC using a two-tailed, unpaired t-test with Welch's correction. Only
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p-values less than 0.05 were considered statistically significant. We used PRISM for all statistical analyses (Version 8.4.0)(455).

Results

We mined published microarray data^{3,4} to identify differentially expressed genes in high-grade serous ovarian cancer (HGSC), the type of ovarian cancer responsible for 70-80% of deaths resulting from the most lethal gynecologic malignancy.

FGF10 is differentially expressed in ovarian tumors from women diagnosed with HGSC.

We identified FGF10 as among the genes whose expression was most different when comparing primary HGSC tumors to normal ovarian tissue² (Table 1). When sorting all of the transcripts measured by microarray based on change in expression between HGSC and the normal ovary, FGF10 ranked 23 out of 29088 total transcripts (Table 1). Differential expression of FGF10 in HGSC tumors was statistically significant (Table 1; $p=9.01E-13$).

We analyzed a second microarray dataset⁴ generated using normal ovarian tissues and tumors from women diagnosed with HGSC to determine whether differential expression of FGF10 could be observed in tumors from a different group of patients. We again found that FGF10 was differentially expressed in primary HGSC tumors (Table 2). When sorting all of the transcripts measured by microarray based on change in expression between HGSC and the normal ovary, in this data set, FGF10 ranked 3909 out of 41093 total transcripts (Table 2). This means the magnitude of change in FGF10 was greater than 90.5% of the measured transcriptome. Differential expression of FGF10 in HGSC tumors in this second microarray dataset was statistically significant (Table 2; $p=8.4E-03$).

FGF10 is expressed at significantly lower levels in HGSC when compared to the normal ovary.

We then obtained exact mRNA expression levels for the differentially expressed FGF10 transcript from both normal ovarian tissue and from high-grade serous ovarian tumors. FGF10 was expressed at significantly lower levels in high-grade serous ovarian cancers when

1 compared to the normal ovary (Figure 1; $p < 0.0001$). We calculated a mean fold change of
2 0.1346 ± 0.0788 in FGF10 expression when comparing HGSC tumors to normal ovarian tissue
3 (Table 1).

4 Thus, we found using published microarray data that FGF10 was among the genes most
5 differentially expressed in tumors from women with high-grade serous carcinomas and that
6 FGF10 expression was significantly lower in HGSC tumors when compared to the normal ovary.
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8 **Discussion**

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10 Transcriptional profiling of HGSC in a Cancer Genome Atlas integrated genomic analysis
11 classified HGSC into four subtypes based on gene clustering: immunoreactive, differentiated,
12 proliferative, and mesenchymal⁷. We sought to continue to describe the transcriptional
13 landscape of high-grade serous ovarian cancers⁷ and identify genes whose differential
14 expression was associated with HGSC by using published microarray data from primary tumors
15 of women diagnosed with HGSC compared to transcriptome data from normal ovarian tissue^{2,3}.
16 In both datasets analyzed, FGF10 was among the genes whose expression changed most
17 significantly when comparing the normal ovary to primary HGSC tumors
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19 Fgf10 is important for the development of the inner ear⁸, otic placode⁹ (a portion of the
20 embryonic ectoderm from which the ear develops), prostate¹⁰, lung^{11,12}, limb¹², pancreas¹³,
21 stomach¹⁴, external genitalia¹⁵, and the cecum¹⁶. Fgf10 is expressed in human ovaries¹⁷. In the
22 buffalo ovary, Fgf10 was specifically expressed in the granulosa cell and theca interna¹⁸. Major
23 phenotypes displayed by Fgf10-deficient mice include cleft palate¹⁹ and congenital heart
24 defects²⁰. Fgf10-deficient embryos do not possess pulmonary veins or arteries, and Fgf10 is
25 required for correct heart positioning in the thoracic cavity²⁰. In the brain, Fgf10-deficient mice
26 manifest delays in differentiation of radial glia in the rostral cortex²¹.
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1 Transgenic mice in which FGF10 was conditionally expressed in the fetal lung under the
2 promoters of surfactant protein or Clara cell secretory protein have disrupted branching
3 morphogenesis and adenomatous malformations, and respiratory failure at birth²². FGF10
4 expression in the lungs after birth lead to the development of benign multifocal pulmonary
5 tumors²².

6 Fgf10 has been labeled as an oncogene, activated by insertional mutagenesis mediated
7 by MMTV in mouse mammary tumors²³. One study documented a role for Fgf10 in cancer of
8 the pancreas, demonstrating expression of Fgf10 from the stroma with expression of the Fgf10-
9 receptor Fgfr2 in pancreatic cancer cells²⁴. In pancreatic cancer cell lines with FGFR2-IIIb
10 expression, Fgf10 was able to induce invasion and migration *in vitro*; this was concomitant with
11 induction of TGF-beta and MT1-MMP in CFPAC-1 cells²⁴. This suggested that soluble signals
12 from the stroma, including Fgf10, were important for programming of invasive and migratory
13 properties in pancreatic cancer cells²⁴. In PTEN-induced models of squamous cell carcinoma,
14 PTEN loss caused significant induction of Fgf10²⁵. Ectopic expression of Fgf10 in the absence
15 of PTEN deletion was sufficient to cause formation of skin papillomas²⁵. There is limited
16 information regarding fibroblast growth factor 10 or its receptors specifically in ovarian cancer.
17 One study documented mutation of FGFR2 in 2.4% of serous ovarian cancers (1 out of 41
18 tumors studied)²⁶.

19 Differential expression of FGF10 in HGSC should be validated in larger and separate
20 cohorts of women with HGSC. If validated, mouse models of high-grade serous ovarian
21 cancers²⁷⁻²⁹ can be utilized to assess the effect of conditional expression of FGF10 in HGSC
22 tumors post-development and on tumor cell proliferation and cancer progression *in vivo*.
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Rank	ID	p-value	t	B	Fold change	Gene	Gene name
23	8111993	9.01E-13	-9.8735364	18.8251135	0.1346 ± 0.0788	FGF10	fibroblast growth factor 10

Table 1: FGF10 is differentially expressed in high-grade serous ovarian carcinomas.

The rank of differential expression, the probe/transcript ID, the p-value with respect to global differential expression, t, a moderated t-statistic, B, the log-odds of differential expression between the two groups compared, the fold change of FGF10 in the patient population when compared to control subjects, the gene and gene name are listed in this chart.

Rank	ID	p-value	t	B	Gene	Gene name
3909	A_32_P66517	8.4E-03	-3.1606701	-2.77316	FGF10	fibroblast growth factor 10

Table 2: FGF10 is among the most differentially expressed genes in a tumor dataset from patients with high-grade serous ovarian carcinomas.

The rank of differential expression, the probe/transcript ID, the p-value with respect to global differential expression, t, a moderated t-statistic, B, the log-odds of differential expression between the two groups compared, the fold change of FGF10 in the patient population when compared to control subjects, the gene and gene name are listed in this chart.

Figure 1: FGF10 is expressed at significantly lower levels in HGSC tumors when compared to the normal ovary.

The mRNA expression of FGF10 in normal ovarian tissues from control subjects (left) and in the primary tumors of patients with HGSC (right) is represented with mean mRNA expression level marked and the result of a statistical test evaluating the significance of difference in FGF10 expression between the ovary of control subjects and primary tumors from patients with HGSC, a p-value, listed above.