Differential expression of SPRY2 in cancer of the vulva.

Shahan Mamoor, MS¹

¹shahanmamoor@gmail.com

East Islip, NY USA

In these brief notes we document work using published microarray data (1, 2) to pioneer integrative transcriptome analysis comparing vulvar carcinoma to its tissue of origin, the vulva. We report the differential expression of sprouty RTK signaling antagonist 2, encoded by SPRY2, in cancer of the vulva. SPRY2 may be of pertinence to understanding transformation and disease progression in vulvar cancer (3).

Keywords: vulvar cancer, vulvar carcinoma, SPRY2, sprouty RTK signaling antagonist 2.

Gynecologic cancers include cervical cancer, ovarian cancer, endometrial (uterine) cancer, and vulvar cancer (4, 3). Vulvar cancer is thought to be the least prevalent of these, with 6190 cases reported in the United States in 2018 (5). We integrated separate published microarray datasets to understand, at the systems level, the transcriptional landscape of vulvar carcinoma.

Methods

We utilized microarray datasets GSE63678 (1) and GSE5563 (2) for this integrated, transcriptome-level differential gene expression analysis of human vulvar cancers in conjunction with GEO2R. GSE63678 was generated using Affymetrix Human Genome U133A 2.0 Array technology with $n=7$ normal vulvar tissues and $n=6$ vulvar carcinoma tissues; analysis was performed using platform GPL571. GSE5563 was generated using Affymetrix Human Genome U133 Plus 2.0 Array technology with $n=10$ normal vulvar tissues and $n=9$ VIN (vulvar intraepithelial neoplasia) tissues; analysis was performed using platform GPL570. The Benjamini and Hochberg method of p -value adjustment was used for ranking of differential expression but raw p -values were used to assess statistical significance of global differential expression. Log-transformation of data was auto-detected, and the NCBI generated category of platform annotation was used. A statistical test was performed to evaluate whether SPRY2 expression was significantly different between normal vulvar tissue and vulvar carcinoma using a two-tailed t -test.

Results

We mined published microarray data (1, 2) to understand the most significant transcriptional changes between the normal vulva and cancer of the vulva.

SPRY2 is differentially expressed in vulvar cancer.

We found that sprouty RTK signaling antagonist 2, encoded by SPRY2, was among the most differentially expressed genes, transcriptome-wide, in cancer of the vulva (1). When sorting each transcript whose expression was measured across the transcriptome based on significance of change between the normal vulva and vulvar cancer tissues, SPRY2 ranked 118 out of 22277 total transcripts, equating to 99.5% differential expression (Chart 1) (1). Differential expression of SPRY2 was statistically significant (Chart 1; $p=1.63E-05$).

We validated our findings by performing a similar analysis using a second published microarray dataset (2), but here measuring gene expression in vulvar intraepithelial neoplasia, a dysplastic cell state from which vulvar carcinomas are said to originate from (6). When sorting each transcript whose expression was measured across the transcriptome based on significance of change between the normal vulva and vulvar cancer tissues, SPRY2 ranked 1033 out of 54662 total transcripts (Chart 2) (2), equating to 98.1% differential expression. Differential expression of SPRY2 was again statistically significant (Chart 2; $p=1.45E-04$).

SPRY2 expression is lower in cancer tissue as compared to the vulva.

We obtained exact mRNA expression data from the first microarray dataset we mined to directly compare expression levels between control and cancer tissues. SPRY2 was expressed at significantly

1 lower levels in cancer of the vulva as compared to normal vulva. SPRY2 was expressed at 8.37 ± 0.59
2 arbitrary units (A.U.) in normal vulva, but at 6.49 ± 0.48 A.U. in vulvar cancer tissues (Figure 1). We
3 calculated a mean fold change of 0.78 in SPRY2 expression when comparing vulvar carcinoma to vulva.

4 **Discussion**

5 We documented here that SPRY2 is differentially expressed at the transcriptome level in cancer and
6 dysplasia of the human vulva (1, 2). SPRY2 is a candidate gene of interest in understanding the
7 transcriptional biology of vulvar cancer.
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28

References

1. Pappa, K.I., Polyzos, A., Jacob-Hirsch, J., Amariglio, N., Vlachos, G.D., Loutradis, D. and Anagnou, N.P., 2015. Profiling of discrete gynecological cancers reveals novel transcriptional modules and common features shared by other cancer types and embryonic stem cells. *PLoS One*, 10(11), p.e0142229.
2. Santegoets, L.A., Baars, R.V., Terlou, A., Heijmans-Antonissen, C., Swagemakers, S.M., van der Spek, P.J., Ewing, P.C., Beurden, M.V., van der Meijden, W.I., Helmerhorst, T.J. and Blok, L.J., 2012. Different DNA damage and cell cycle checkpoint control in low- and high-risk human papillomavirus infections of the vulva. *International journal of cancer*, 130(12), pp.2874-2885.
3. Canavan, T.P. and Cohen, D., 2002. Vulvar cancer. *American family physician*, 66(7), p.1269.
4. Grimes, D.A. and Economy, K.E., 1995. Primary prevention of gynecologic cancers. *American journal of obstetrics and gynecology*, 172(1), pp.227-235.
5. Weinberg, D. and Gomez-Martinez, R.A., 2019. Vulvar cancer. *Obstetrics and Gynecology Clinics*, 46(1), pp.125-135.
6. Del Pino, M., Rodriguez-Carunchio, L. and Ordi, J., 2013. Pathways of vulvar intraepithelial neoplasia and squamous cell carcinoma. *Histopathology*, 62(1), pp.161-175.

Rank: 118
Probe ID: 204011_at
p-value: 1.63E-05
t: 6.526986
B: 3.32472
Gene: SPRY2
Gene name: sprouty RTK signaling antagonist 2

Chart 1: Differential expression of SPRY2 in vulvar cancer.

Rank of differential expression, probe ID, *p*-value of global differential expression, *t*, a moderated *t*-statistic, *B*, the log-odds of differential expression between the groups compared, gene and gene name are listed in this chart.

Rank: 1033
Probe ID: 204011_at
p-value: 1.45E-04
t: 4.7038678
B: 1.044802
Gene: SPRY2
Gene name: sprouty RTK signaling antagonist 2

Chart 2: Differential expression of SPRY2 in VIN tissues.

Rank of differential expression, probe ID, *p*-value of global differential expression, *t*, a moderated *t*-statistic, *B*, the log-odds of differential expression between the groups compared, gene and gene name are listed in this chart.

Figure 1: SPRY2 is expressed at significantly lower levels in vulvar cancer as compared to normal vulva.

The mRNA expression level of SPRY2 in the normal vulva (left) and in vulvar carcinoma (right) is graphically depicted with the result of a statistical test evaluating significance of difference in SPRY2 expression between the two groups, a *p*-value, listed above.