HPV-induced cervical, oropharyngeal and anal carcinoma share theranostic biomarkers involved in immunomodulation and PIK3CA signal transduction pathway.

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Abstract (No. 11107)

Background: Several distinct cancers are caused by human papillomavirus (HPV)-including squamous cell carcinomas of the cervix (CSCC), anal canal (ASCC), and oropharynx (OSCC). Importantly, platinum based chemoradiation protocols are similarly effective in these entities. We assessed CSCC, ASCC, and OSCC for additional evidence of shared characteristics which could be used to identify potential molecular targets across the spectrum of HPV induced cancers.

Methods: 201 ASCC, 321 CSC and 358 OSCC tumors underwent molecular profiling with a multiplatform approach (Caris Life Sciences). TP53 wild type status was used as a surrogate for HPV. Testing included sequencing (NGS, Sanger), protein expression (IHC) and gene amplification (ISH). The 2-tail Fisher's exact test ($p \leq 0.05$), JMPv10.0 (SAS Institute Inc., Cary, NC) was utilized for statistical analysis.

Results: Excluding TP53-mutated patients, 197 ASCC, 317 CSCC and 317 OSCC were included in the study. Multiplatform profiling reveals marked similarities among the HPV-induced cancers. None of the frequencies observed displayed statistically significant differences. Selected results appear below.

Frequency (positivity, underexpression#, amplified or mutated) – greyed boxes indicate Rx is NCCN-endorsed				
	Rx Association	Anal	Cervical	Oropharyngeal
PGP IHC	Taxanes	9%	3%	3%
TLE3 IHC		32%	31%	37%
TUBB3# IHC		89%	75%	83%
ERCC1# IHC	Platinums	51%	60%	67%
BRCA1 NGS		10%	15%	7%
BRCA2 NGS		10%	30%	10%
EGFR IHC	Anti-EGFR	92%	70%	92%
EGFR_ISH		7%	17%	18%
HER2	Anti-HER2	0%	4%	2%
HER2_ISH		2%	6%	3%
PTEN# IHC	PAM pathway inhibitors	55%	47%	50%
PIK3CA NGS		29%	34%	18%
PTEN NGS		4%	2%	5%
AKT NGS		2%	2%	4%
PD1 IHC	Immuno-modulatory	50%	78%	81%
PDL1 IHC		0%	18%	9%
FBXW7 NGS	mTOR inhibitors	14%	6%	6%
KRAS NGS	MEK inhibitors	1%	3%	4%

Conclusions: Unlike the genomic instability observed in many solid tumors, HPV-induced carcinogenesis yields a more homogenous phenotype. These data support previous work identifying the PIK3CA-AKT-mTOR pathway as a potential target. Given the phenomenon of HPV E6 & E7-induced oncogene addiction following viral integration, the need to explore PD1/PDL1 inhibition in these cancers is implicit.

Background

- Persistent infection with oncogenic HPV subtypes is etiologically linked to cervical (virtually all), oropharyngeal (70%) and anal (90%) cancers, among others.
- The critical molecules that facilitate malignant transformation are viral oncoproteins, E6 & E7, which interact TP53 and pRB, to seize cell cycle control and promote proliferation. TP53 mutation is inversely correlated with HPV status.
- During viral persistence, it is speculated that virally infected cells evade and may actively suppress the induction of a positive immune response. Immunomodulatory therapies, therefore, are actively being investigated for cancers that are in a state of immune tolerance (i.e. virally-driven)
- Currently, platinum-based radiation protocols are the only unifying treatment approach across HPV-induced cancers.

Methods

ASCC, CSCC and OSCC patients whose tumors were sent to Caris Life Sciences for molecular profiling were analyzed retrospectively for patterns of theranostic biomarker expression. TP53 wildtype status was used as a surrogate for HPV. Testing included sequencing (NGS or Sanger, up to 47 genes assayed), protein expression (IHC, up to 23 genes assayed) and gene amplification (ISH, up to 5 genes assayed). Fisher's exact test ($p<0.05$), JMPv.10.0 (SAS Institute Inc., Cary, NC) was utilized for statistical analysis.

Results

HPV cancer type	n	Sex		Age	Disease state	
		male	female	average (range)	Primary	Metastatic*
ASCC	197	37%	63%	59 (31-89)	25%	75%
CSCC	310	0%	100%	49 (22-89)	40%	60%
OSCC	317	77%	23%	57 (21-90)	47%	53%

Table 1. Clinicopathologic Features of Molecularly Profiled HPV-induced Cancers. OSCC was dominated by the male sex (77%) and ASCC, by the female sex (63%). CSCC exhibited the lowest mean age (49), followed by OSCC and ASCC (57 & 59, respectively). Amongst tumors profiled, 75% of ASCC were metastatic specimens, followed by 60% of CSCC and 53% OSCC.

Metastatic Sites	ASCC	CSCC	OSCC
Abdomen, NOS	0%	6%	1%
Bone	0%	1%	4%
Breast	0%	1%	0%
Cervix	1%	0%	0%
Chest, NOS	0%	0%	3%
CNS	3%	0%	1%
Colon	3%	2%	0%
Connective & Soft Tissue	11%	7%	24%
Intra-abdominal site, NOS	1%	0%	0%
Liver	18%	4%	7%
Lung & Bronchus	6%	9%	21%

Metastatic Sites	ASCC	CSCC	OSCC
Lymph nodes	21%	20%	18%
Mediastinum	1%	0%	1%
Omentum	1%	2%	0%
Other female	0%	1%	0%
Other parts of tongue	0%	0%	4%
Oral Cavity, Mouth, NOS	0%	0%	5%
Pancreas	0%	1%	0%
Pelvis, NOS	4%	12%	0%
Penis & Scrotum	2%	0%	0%
Pharynx/Nasopharynx	0%	0%	3%
Pleura	0%	1%	1%

Metastatic Sites	ASCC	CSCC	OSCC
Rectum	17%	1%	0%
Retroperitoneum & peritoneum	1%	5%	0%
Sacrum	1%	0%	0%
Skin	3%	1%	6%
Small Bowel	0%	0%	1%
Urinary Bladder	1%	2%	0%
Uterus, NOS	0%	11%	0%
Vagina & Labia	3%	12%	0%
Vulva, Nos	1%	1%	0%

Table 2. Frequency of *Metastatic sites profiled. All three HPV-induced cancers exhibited frequent metastases to lymph nodes. In addition to lymph nodes, the most frequent metastatic sites profiled was liver (18%) for ASCC, pelvis, nos and vagina/labia (both 12%) for CSCC and connective/soft tissues (24%) for OSCC.

Results, continued

Drug Type	Predictive Biomarkers	HPV-induced Tumor Type		
		ASCC (n=198)	CSCC (n=310)	OSCC (n=317)
Cytotoxic Therapies	ERCC1, BRCA	cisplatin	cisplatin, carboplatin	cisplatin, carboplatin
	TS	5-fluorouracil, capecitabine	5-fluorouracil, pemetrexed	5-fluorouracil, capecitabine
	TUBB3/TLE3/PGP		paclitaxel, docetaxel, vinorelbine	paclitaxel, docetaxel
	TOPO1		topotecan, irinotecan	
	RRM1		gemcitabine	
	BRCA1/2	mitomycin	mitomycin	
Targeted Therapies	none identified		bevacizumab	cetuximab

Table 3. Comparison of cancer agents according to NCCN guidelines for HPV-induced Cancers. ASCC has the least number of agents (those with predictive biomarkers) recognized by NCCN, including a complete lack of approved targeted therapies. CSCC has the greatest number of agents to choose from. Both CSCC and OSCC have targeted agents available, however predictive biomarkers have not been identified.

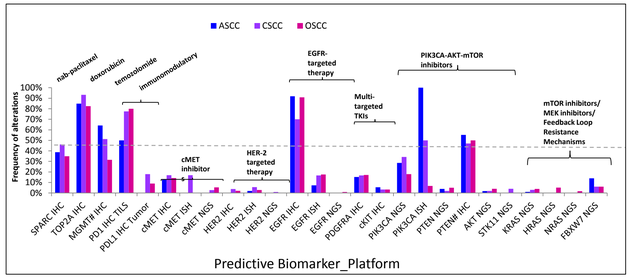

Figure 2 – Off-Compendium, Targetable alterations identified by IHC, ISH and NGS. (% overexpressed, or amplified; # indicates biomarkers where low or no expression is predictive of therapy response). Dotted line indicates biomarkers with frequency of alterations effecting at least half of the population tested: TOP2A, MGMT, **PD1**, EGFR and **PIK3CA**. Treatment strategies targeting these proteins should be prioritized based on potential for impact. (ASCC: n=184 for IHC, n=100 for ISH, n=95 for sequencing; CSCC: n=299 for IHC, n=178 for ISH, n=241 for sequencing; OSCC: n=306 for IHC, n=171 for ISH, n=153 for sequencing).

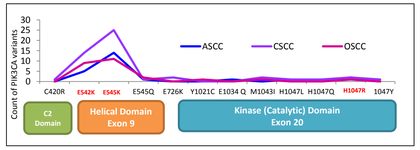

Figure 3 – Hot spot mutations (E542K, E545K and H1047R) account for 83% of the variants detected in PIK3CA, across HPV-induced cancers. Exon 9 variants, in the helical domain predominate over exon 20 mutations. 35% (6/17) ASCC PIK3CA mutants co-occur with FBXW7 vs. 5% (2/37) for CSCC and 6% (1/17) for OSCC. 75% (3/4) HPV-induced cancers w/ PIK3CA amplification exhibit simultaneous PIK3CA mutation. A minority (2%) of ASCC, CSCC and OSCC exhibit PIK3CA + KRAS mutations. Implications for targeting HPV-induced cancers with helical domain mutations, co-mutations with FBXW7 or additional oncogenic events in the PIK3CA pathway is under clinical investigation.

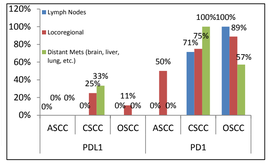

Figure 4 – Expression of PD1 and PDL1 immune checkpoint biomarkers by IHC, according to site of specimen used for profiling. Uniform patterns of PD1 in tumor infiltrating lymphocytes (TILs) and PDL1 in tumor cells, for HPV-induced cancers is lacking. Highest PDL1 expression is observed in distant metastases of CSCC, and highest PD1 expression is observed in lymph nodes samples of OSCC and distant metastases of CSCC.

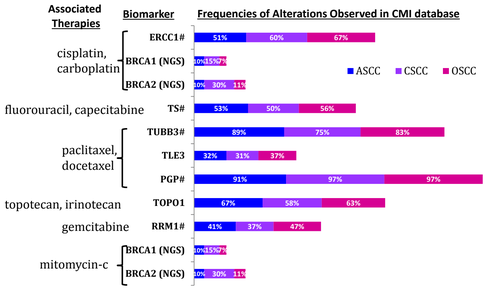

Figure 1. Comparison of biomarker frequencies for NCCN-endorsed therapy recommendations for HPV-induced cancers. Similar frequencies of expression are suggestive that therapies may be effective across tumor types. (ASCC: n=184 for IHC, n=95 for sequencing; CSCC: n=299 for IHC, n=241 for sequencing; OSCC: n=306 for IHC, n=153 for sequencing).

Conclusions

- This is one of the first studies to explore biomarker expression in a spectrum of HPV-induced carcinomas.
- With widespread adoption of platinum-based chemoradiation protocols, acquired drug resistance via ERCC1 may limit the efficacy of platinum-based regimens at the time of recurrence.
- While anti-EGF strategies may be limited by lack of a suitable prognostic biomarker, the PI3K3CA-AKT-mTOR signal transduction pathway may harbor both predictive and therapeutic biomarkers which could be exploited for therapeutic gain using dual PI3K and/or mTOR inhibitors.
- The phenomenon of virally (ie., E6 and E7) driven oncogene addiction in these carcinomas indicates that checkpoint inhibition (eg., PD1/PDL1 and possibly CTLA4) may represent a valid immunotherapeutic strategy that may confer a survival advantage among patients with advanced disease.
- Small, but significant incidence of BRCA1/2 across these tumor types, may point to greater benefit to platinum compounds, mitomycin or even PARP inhibitors.
- These data demonstrate these HPV-induced cancers share similar molecular alterations, which justifies the molecular basis for thinking of these tumors together (and putting them under same umbrella when planning clinical trials).

References

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