Genomic Landscape of Angiosarcoma: A Targeted and Immunotherapy Biomarker Analysis of 143 Patients.

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Background

Angiosarcoma (AS) is an uncommon highly aggressive sarcoma that exhibits endothelial differentiation and represents less than 1% of all STS, with an annual incidence of 300 cases in the US. In locally advanced or metastatic disease, initial responses to cytotoxic chemotherapy are common; however, the duration is limited, and most patients will eventually succumb to metastatic disease. Common regimens used include weekly paclitaxel, gemcitabine + docetaxel and immunomodulatory agents such as steroids + cyclophosphamide. Unfortunately, once metastasized, median OS is only 3 to 12 months. Targeted therapies for AS including VEGF blocking have limited efficacy and the predictive value of recurrent genetic alterations is still not well characterized.

More recently, we've seen reports of exceptional responses with use of Immunotherapy (IO) in some AS patients[1,2]. However, predictor factors for IO-response in sarcoma are in an early stage of development. Understanding distinct molecular drivers as well as immune microenvironment appears to be crucial to predict these responses[3]. Herein, we performed molecular analyses to identify new, potential treatment options including immunotherapy for patients with AS.

Methods:

- Retrospectively reviewed 143 AS tumors profiled with Caris Life Sciences from 2015-2019
- Next-Generation Sequencing (NGS) using the NextSeq platform and a custom-designed SureSelect XT assay to enrich 592 cancer-related whole-gene targets was performed on each tumor.
- Whole transcriptome sequencing (WTS) was performed on 53 tumors and used for microenvironment cell population (MCP)-counter analysis.
- Biomarkers potentially associated with response to IO-therapy (TMB-High [≥10/Mb], MSI-High, and PD-L1 [IHC ≥ 2+ and 5%]) were also analyzed.
  - TMB = Tumor Mutational Burden; MSI = Microsatellite Instability
- Molecular results were evaluated according to primary tumor site
- Statistical analysis was performed using Chi-square or Fisher's exact tests where appropriate (*raw p<0.05; **q<0.05 Benjamini & Hochberg adjusted p-value for multiple hypothesis testing)

Results:

Most common pathogenic mutations: TP53 (29.2%), MYC amp (23%), ARID1A (17.1%), POT1 (17.7%), and ATRX (13.2%). Figure 1.

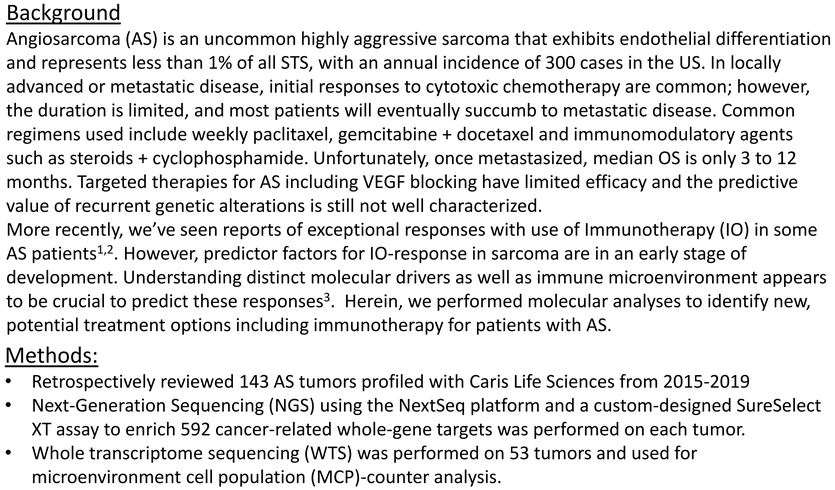

Figure 1. Oncoprint for Most Frequently Altered Biomarkers.

Molecular biology of angiosarcoma varies according to primary site.

Head and Neck AS:

- **IO-response markers:** 65% of cases (n=28/43; p<0.0001, q=0.0149). Figure 3B.
  - ➢ TMB-High observed in 62.5% (n=25/40; p<0.0001, q<0.0001). Figure 2.
- TP53 mutation present in 50.0% (n=21/42; p=0.0004, q=0.0716). Figure 3A.
- POT1 mutation present in 40.5% (n=17/42; p<0.0001, q=0.0044). Figure 3A.
- ARID1A mutation present in 33.3% (n=5/15; p=0.5875, q=1.0). Figure 3A.

Breast AS:

- Cell cycle pathway aberrations were the most common (Figure 3B) and almost entirely driven by MYC amplification present in 63.3% (n=19/30; p<0.0001, q<0.0001).
- HRAS mutations present in 16.1% (n=5/31; n=p=0.0377, q=1.0). Figure 3A.
- PI3KCA mutation present in 16.1% (n=5/31; n=p=0.2352, q=1.0). Figure 3A.
  - ➢ Highest among AS subgroups.

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Total (N)	Breast (N)	Extremity (N)	Head & Neck (N)	Cutaneous (N) ⁜	Visceral (N)	Other (N)
143	31	15	43	9	30	15

Table 1. Stratification of patients based on primary AS site. ⁜ Cutaneous locations other than head and neck, breast or extremity.

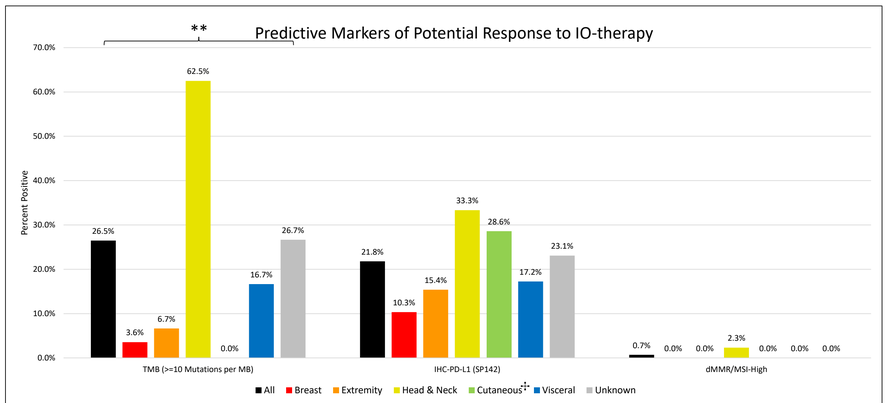
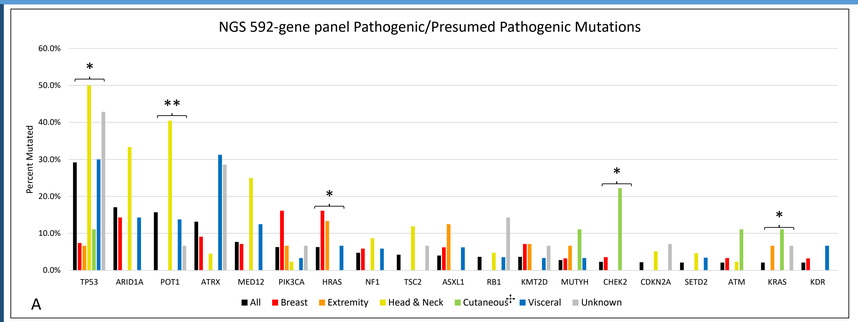


Figure 2. Frequency of predictive markers of potential response to IO-therapy by primary AS site. ** P-value and Q-value < 0.05. ⁜ Cutaneous locations other than head and neck, breast or extremity.



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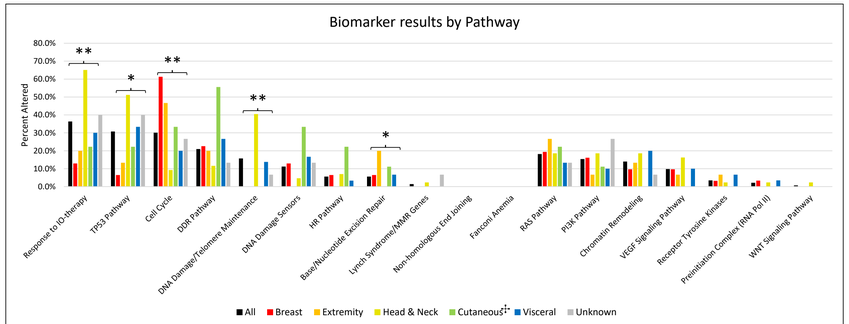

Figure 3. Frequency of biomarkers stratified by primary site. A. Specific biomarker alteration frequencies (Biomarkers mutated in ≥ 3 cases were included). B. Frequencies of mutated genes grouped by pathway.
** P-value and Q-value < 0.05, * P-value < 0.5, but Q-value > 0.05. ⁜ Cutaneous locations other than head and neck, breast or extremity.

MCP-counter:

- Total cohort divided in 4 distinct immune classes:
  1. Immune-High – B lineage high (13.2%)
  2. Vascularized – Endothelial cells high (24.5%)
  3. Immune-Desert (41.5%)
  4. Heterogeneous – Moderate abundance (20.8%)
  - ➢ Immune class signatures were evenly distributed among different primary sites.
- Interestingly, the Immune-High class had the lowest median TMB = 6 muts/MB (range 3-17).

Future Directions for Research:

- Further prospective clinical trials for the use of IO-therapy in Head and Neck AS are warranted.
- The use of MCP-counter as an adjunctive predictive method needs validation in prospective AS trials.
- Interestingly, Breast AS was enriched for cell cycle pathway aberrations. Consequently, prospective trials targeting cell cycle regulators in Breast AS need to be explored.
- Truncating mutations in ARID1A (which codes for part of SNF/SWI complex) were present in 17% of our cohort. We need to further explore use of EZH2 inhibitors in this scenario.

References:

1. Florou, Vaia, Andrew E Rosenberg, Eric Wieder, Krishna V Komanduri, Despina Kolonias, Mohamed Uduman, John C Castle, Jennifer S Buell, Jonathan C Trent, and Breelyn A Wilky. 2019. "Angiosarcoma Patients Treated with Immune Checkpoint Inhibitors : A Case Series of Seven Patients from a Single Institution"JITC 7: 1–8. https://doi.org/10.1186/s40425-019-0689-7.
2. Painter, Corrie A., Esha Jain, Brett N. Tomson, Michael Dunphy, Rachel E. Stoddard, Beena S. Thomas, Alyssa L. Damon, et al. 2020. "The Angiosarcoma Project: Enabling Genomic and Clinical Discoveries in a Rare Cancer through Patient-Partnered Research." *Nature Medicine* 26 (2): 181–87. https://doi.org/10.1038/s41591-019-0749-z.
3. Petitprez, Florent, Aurélien de Reyniès, Emily Z. Keung, Tom Wei Wu Chen, Cheng Ming Sun, Julien Calderaro, Yung Ming Jeng, et al. 2020. "B Cells Are Associated with Survival and Immunotherapy Response in Sarcoma." *Nature* 577 (7791): 556–60. https://doi.org/10.1038/s41586-019-1906-8.