GNAS Driver Mutations in Primary and Metastatic Nonmucinous Appendiceal Adenocarcinoma Versus Analogous Colorectal Cancers. An AACR Project GENIE Analysis.

Andrew M. Fleming MD[1,2], Leah E. Hendrick MD MS[1], Julia Pedo Freitas MD[1], Suraj Sarvode Mothi MPH[3], Ian B. Solsky MD[1], Danny Yakoub MD PhD[4], Andrew J. Murphy MD[1,2], Elizabeth Gleeson MD MPH[5], Evan S. Glazer MD PhD[1], David Shibata MD[1], Jeremiah L. Deneve DO[5]

[1] Department of Surgery, The University of Tennessee Health Science Center, Memphis, TN, USA
[2] Department of Surgery, St. Jude Children's Research Hospital, Memphis, TN, USA
[3] Department of Biostatistics, St. Jude Children's Research Hospital, Memphis, TN, USA
[4] Division of Surgical Oncology, Augusta University Medical Center, Augusta, GA, USA
[5] Department of Surgery, University of North Carolina, Chapel Hill, NC, USA

Corresponding Author: Jeremiah L. Deneve DO
Department of Surgery
University of North Carolina
170 Manning Dr
Physicians Office Building, Room 1147, CB#7213
Chapel Hill, NC 27599-7213
Phone: 919-966-5221
Email: jdeneve@unc.edu

The authors included each contributed substantially to this research as per the guidelines of the International Committee of Medical Journal Editors (ICMJE).

Disclosures: The authors declare no financial disclosures or conflicts of interest related to the research described in this manuscript.

Acknowledgements: The authors would like to acknowledge the American Association for Cancer Research and its financial and material support in the development of the AACR Project GENIE registry, as well as members of the consortium for their commitment to data sharing. Interpretations are the responsibility of study authors.

Background
Systemic therapy recommendations for appendiceal adenocarcinoma (AA) are based on data from colorectal cancer (CRC) studies. A more nuanced understanding of the genomic underpinnings of AA could help drive the development of more precise and effective systemic therapies. The current study assessed distinct driver mutations in primary and metastatic AA and CRC using the largest publicly accessible cancer clinicogenomic dataset.

Methods
The American Association for Cancer Research (AACR) Project Genomics Evidence Neoplasia Information Exchange (GENIE) dataset "GENIE Cohort v14.0-public," released September 2023, was queried for CRC and AA samples via cBioPortal for Cancer Genomics. Exclusion criteria included non-adenocarcinoma histology, mucinous histology, and missing data regarding primary versus metastatic site of origin. Sample-level genomic alterations were compared using Fisher's exact tests, with multiple hypothesis testing corrected via the Benjamini-Hochberg method. q-values <.01 denoted statistical significance. Volcano plots of log2 alteration ratios and -log10 p-values were used to visualize differential rates of mutations between groups.

Results
15,117 CRC samples (10,056 primary, 5,061 metastatic) and 300 AA samples (133 primary, 167 metastatic) were included. In the primary samples, there were no differences between groups regarding age, sex, or race, (all q>.05). In the metastatic samples, patients with AA were more often female (59.3% vs. 46.0%, q<.01) than those with mCRC, but similar regarding age and race (both q>.05). When compared to primary CRC, primary AA had higher rates of mutations in *GNAS* (24.2% vs. 4.8%, q<.0001). Metastatic AA also had higher rates of *GNAS* mutations than metastatic CRC (15.7% vs. 4.5%, q<.0001) (Figure 1A-B). Among *GNAS* mutations in metastatic AA, *R201H* (9.2% vs. 0.8%, q<.0001) and *R201C* (6.1% vs. 0.4%, q<.0001) missense putative driver mutations in the switch I domain on exon 8 of the GNAS protein were more commonly identified than in metastatic CRC. *APC* mutations were less common in AA than CRC in both primary (13.7% vs. 67.0%, q<.0001) and metastatic (8.4% vs. 66.4%, q<.0001) samples. *TP53* mutations were also less common in both primary AA (39.4% vs. 68.7%, q<.0001) and metastatic AA (34.0% vs. 73.1%, q<.0001) than in analogous CRC samples.

Conclusions
Higher rates of oncogenic *GNAS* mutations are observed in AA than CRC in both the primary and metastatic setting, even when excluding mucinous histologies. Investigating *GNAS* as a potential therapeutic target in patients with $GNAS^{R201H}$ and $GNAS^{R201C}$-mutant AA should be prioritized.

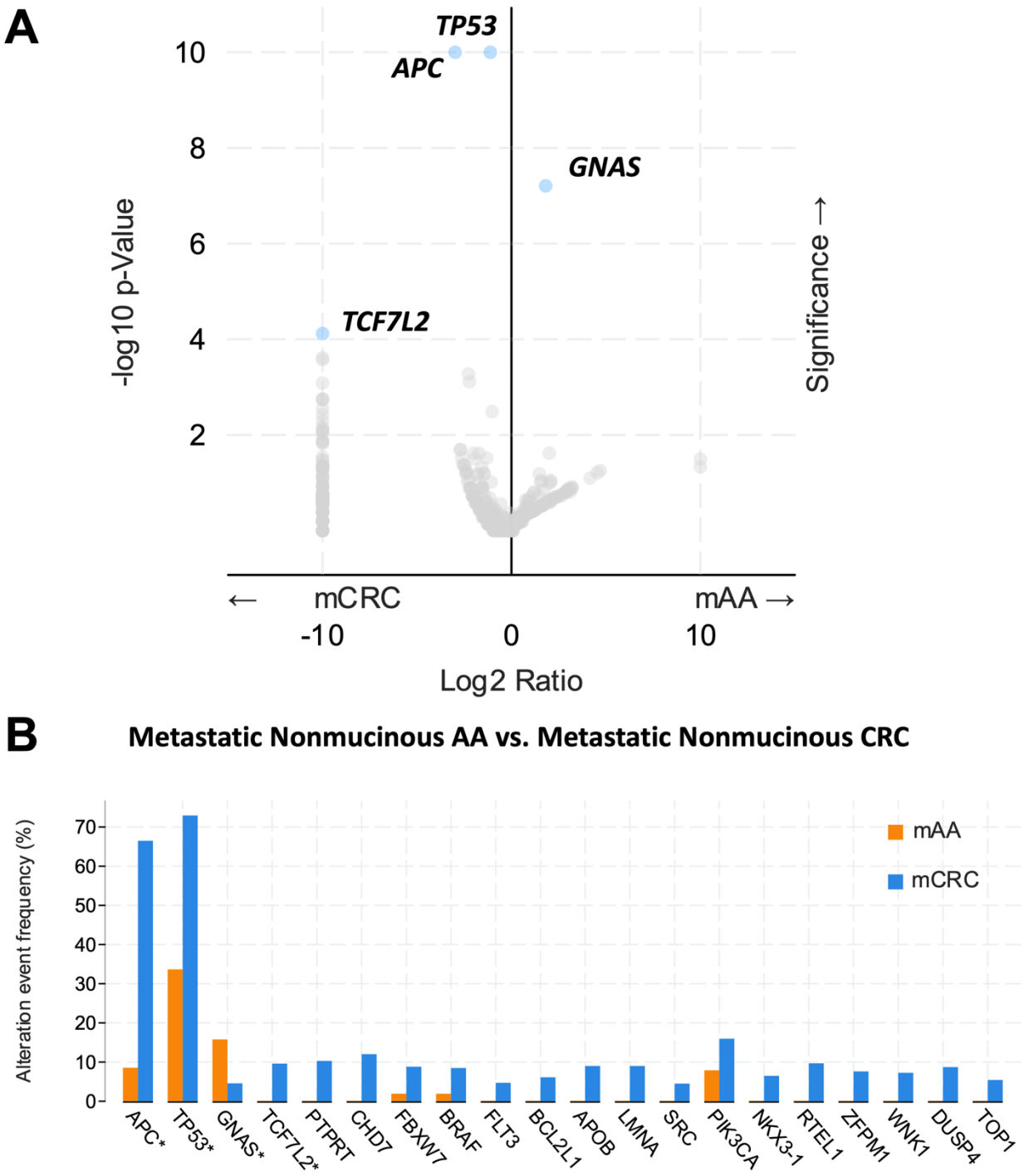

Figure 1. Comparison of mutations profiled in metastatic nonmucinous appendiceal adenocarcinoma and metastatic nonmucinous colorectal adenocarcinoma. **(A)** Volcano plot of Log2 genetic alteration ratios and -log10 P values demonstrating significantly higher rates of *GNAS* mutations in metastatic nonmucinous appendiceal carcinoma. **(B)** Bar graph demonstrating significantly higher rates of *GNAS* mutations in metastatic nonmucinous appendiceal carcinoma.