DR. JOHN MCCOY (Orcid ID : 0000-0003-1577-9910)
DR. CARLOS WAMBIER (Orcid ID : 0000-0002-4636-4489)
DR. SERGIO VANO-GALVAN (Orcid ID : 0000-0003-2773-7494)
DR. ANDY GOREN (Orcid ID : 0000-0002-8190-2289)

Article type : Letter to Editor

Androgen Receptor Genetic Variant Predicts COVID-19 Disease Severity: A Prospective Longitudinal Study of Hospitalized COVID-19 Male Patients

John McCoy, PhD_1*, Carlos Gustavo Wambier, MD, PhD_2, Sabina Herrera, MD, PhD_3, Sergio Vaño-Galván, MD, PhD_4, Francesca Gioia, MD_3, Belen Comeche, MD_3, Raquel Ron, MD_3, Sergio Serrano-Villar, MD, PhD_3, Rafal M Iwasiow, PhD_5, Michael A Tayeb, MSc_5, Flávio Adsuara Cadegiani, MD, PhD_6, Natasha Atanaskova Mesinkovska, MD, PhD_7, Jerry Shapiro, MD_8, Rodney Sinclair, MD, PhD_9, Andy Goren, MD_1.

$_1$Applied Biology, Inc. Irvine, CA, USA.
$_2$Department of Dermatology, Alpert Medical School of Brown University, Providence, RI, USA.
$_3$Infectious Diseases Unit, Ramón y Cajal Hospital, Madrid, Spain.
$_4$Dermatology Department, Ramón y Cajal Hospital, Madrid, Spain.
$_5$DNA Genotek Inc. 3000-500 Palladium Drive, Ottawa, ON K2V 1C2 Canada
$_6$Department of Clinical Endocrinology, Federal University of São Paulo Medical School, Sao Paulo, Brazil.
$_7$Department of Dermatology, University of Claifornia, Irvine, CA, USA.
$_8$Ronald O. Perelman Department of Dermatology, New York University School of Medicine, NY, USA.
$_9$Sinclair Dermatology, Melbourne, VIC, Australia.

***Corresponding author:**
John McCoy, PhD
Applied Biology, Inc.
17780 Fitch, Suite 192
Irvine, CA 92614
johnm@appliedbiology.com

Funding sources: None

Conflicts of Interest: None declared.

IRB approval status: The study was approved by the ethics committee at Ramon y Cajal Hospital.

Manuscript word count: 600 words
References: 6
Figures: 0
Supplementary figures: 0 Tables: 0 Supplementary tables: 0

Keywords: COVID-19; SARS-CoV-2; androgen receptor; androgenetic alopecia; anti-androgen therapy; transmembrane protease serine 2; TMPRSS2; Enzalutamide, Dutasteride; Finasteride; 5-alpha reductase; Spironolactone;

Abbreviations:

5ARi: 5-alpha reductase inhibitors

ICU: Intensive care unit

TMPRSS2: Transmembrane protease, serine 2

AA: Androgenetic alopecia

Disclosure Statements:

John McCoy, PhD: Dr. McCoy has nothing to disclose.

Carlos Gustavo Wambier, MD, PhD: Dr. Wambier has nothing to disclose.

Sabina Herrera, MD, PhD: Dr. Herrera has nothing to disclose.

Sergio Vaño-Galván, MD, PhD: Dr. Vano-Galvan has nothing to disclose.

Francesca Gioia, MD: Dr. Gioia has nothing to disclose.

Belen Comeche, MD: Dr. Comeche has nothing to disclose.

Raquel Ron, MD: Dr. Ron has nothing to disclose.

Sergio Serrano-Villar, MD, PhD: Dr. Serrano-Villar reports grants, personal fees and non-financial support from Gilead Sciences, grants, personal fees and non-financial support from Merck, Sharp and Dohme, personal fees and non-financial support from ViiV Healthcare, personal fees from Janssen, outside the submitted work.

Rafal M Iwasiow, PhD: Dr. Iwasiow reports personal fees from DNA Genotek, Inc. outside the submitted work.

Michael A Tayeb, MSc: Mr. Tayeb reports personal fees from DNA Genotek, Inc. outside the submitted work.

Flávio Adsuara Cadegiani, MD, PhD: Dr. Cadegiani has nothing to disclose.

Natasha Atanaskova Mesinkovska, MD, PhD: Dr. Mesinkovska has nothing to disclose.

Jerry Shapiro, MD: Dr. Shapiro has nothing to disclose.

Rodney Sinclair, MD, PhD: Dr. Sinclair has nothing to disclose.

Andy Goren, MD: Dr. Goren has nothing to disclose.

To the Editor, Men infected with SARS-CoV-2 are more likely to be admitted to the intensive care unit (ICU) compared to women.[1] Previously, we have reported that among hospitalized men with COVID-19, 79% presented with androgenetic alopecia (AA) compared to 31-53% that would be expected in a similar aged match population.[2] AA is known to be mediated by variations in the androgen receptor (AR) gene.[3] In addition, the only known promoter of the enzyme implicated in SARS-CoV-2 infectivity, TMPRSS2, is regulated by an androgen response element.[4] The polyglutamine repeat (CAG repeat) located in the AR gene is associated with androgen sensitivity and AA.[3] These observations led us to hypothesize that variations in the AR gene may predispose male COVID-19 patients to increased disease severity.

We conducted a prospective longitudinal study of hospitalized COVID-19 males. The subjects were categorized into two cohorts: subjects with a CAG>=22 and subjects with a CAG<22. Subjects taking androgen modifying drugs, e.g., 5ARis, were excluded. DNA was collected using ORAcollect•Dx: (DNAGenotek, Ottawa, Canada). AR CAG repeat region was PCR-amplified and 300bp paired-end sequencing was performed using a MiSeq (Illumina, San Diego, California). Reads were mapped to reference AR sequences containing 1 to 50 CAG repeats, the reference with the greatest number of mapped reads was reported as the CAG repeat count. Subjects were followed for a period of 60 days from the date of hospitalization. Primary and secondary outcomes were the rate of ICU admissions and length of hospitalization, respectively.

77 COVID-19 positive men were recruited to the study; 12 were excluded due to their use of androgen modifying drugs, leaving 65 patients enrolled in the study. 31 (48%) subjects had a CAG<22, with average age of 67.9 (+/- 12.3). The median duration of hospitalization among subjects with a CAG<22 was 25 days (95% CI: 9.000-41.6512), and 14 (45.2%) were admitted to the ICU. 34 (52%) subjects had a CAG>=22, their average age was 65.0 (+/- 12.15). Among the 34 subjects with a CAG>=22, the median duration of hospitalization was 47.5 days (95% CI: 22.9533-49.0935), and 24 (70.6%) were admitted to the ICU.

The proportion of subjects admitted to the ICU with CAG<22 was significantly lower than the proportion of subjects with CAG>=22 (Fisher's exact test p= 0.046791. Subjects with a CAG>=22 had a higher risk for ICU admissions compared to subjects with a CAG<22: OR 2.9143(95% CI: 1.0487-

8.0985) and Likelihood Ratio 1.705(95% CI: 0.985-2.951). Further, estimating 40% of hospitalized COVID-19 male patients are likely admitted to the ICU,[5] the Bayes' adjusted positive predictive value of the AR CAG score in predicting ICU admissions was 53.202% (95%CI: 39.646%-66.301%) and the negative predictive value was 71.938% (95%CI: 60.693%-80.974%).

Our data suggest that longer AR CAG score is associated with more severe COVID-19 disease. In some androgen mediated disease, short CAG has been associate with worse prognosis, e.g., in prostate cancer.[6] However, in skeletal muscle, a long CAG repeat length produces higher androgen mediated activity.[7] We believe this discrepancy can be explained by the tissue dependent expression of co-factors important for activation of the androgen response element (ARE).[8] For example, protein arginine methyltransferase 6 has been shown to be highly expressed in lung and has been shown to be a specific co-activator of the androgen receptor.[9]

The results of this study suggest that the AR CAG repeat length could potentially be used as a biomarker to identify male COVIID-19 patients at risk for ICU admissions. More importantly, identification of a biomarker associated with the androgen receptor is yet another piece of evidence supporting the important role of androgens in SARS-CoV-2 disease severity. We recognize the limitations of this small study; however, our findings, combined with previous reports implicating androgens in COVID-19 disease severity,[3-5] should encourage other groups to explore interventional studies of anti-androgens in COVID-19 patients. Currently, we are conducting a double-blinded interventional study with dutasteride (NCT04446429).

REFERENCES

1. Wadman M. Sex hormones signal why virus hits men harder. *Science (80-)*. 2020;368(6495):1038-1039. doi:10.1126/science.368.6495.1038
2. Gebhard C, Regitz-Zagrosek V, Neuhauser HK, Morgan R, Klein SL. Impact of sex and gender on COVID-19 outcomes in Europe. *Biol Sex Differ*. 2020;11(1):29. doi:10.1186/s13293-020-00304-9
3. Wambier CG, Goren A, Vaño-Galván S, et al. Androgen sensitivity gateway to COVID-19 disease severity. *Drug Dev Res*. May 2020:ddr.21688. doi:10.1002/ddr.21688
4. Wambier CG, Vaño-Galván S, McCoy J, et al. Androgenetic Alopecia Present in the Majority of Hospitalized COVID-19 Patients – the "Gabrin sign." *J Am Acad Dermatol*. May 2020. doi:10.1016/j.jaad.2020.05.079
5. Montopoli M, Zumerle S, Vettor R, et al. Androgen-deprivation therapies for prostate cancer and risk of infection by SARS-CoV-2: a population-based study (N = 4532). *Ann Oncol*. May 2020. doi:10.1016/j.annonc.2020.04.479
6. Tirabassi G, Cignarelli A, Perrini S, et al. Influence of CAG Repeat Polymorphism on the Targets of Testosterone Action. *Int J Endocrinol.* 2015;2015:298107. doi:10.1155/2015/298107
7. Sheppard RL, Spangenburg EE, Chin ER, Roth SM. Androgen receptor polyglutamine repeat length affects receptor activity and C2C12 cell development. Physiol Genomics. 2011;43(20):1135-1143. doi:10.1152/physiolgenomics.00049.2011
8. Heemers HV, Tindall DJ. Androgen receptor (AR) coregulators: a diversity of functions converging on and regulating the AR transcriptional complex. Endocr Rev. 2007;28(7):778-808. doi:10.1210/er.2007-0019
9. Scaramuzzino C, Casci I, Parodi S, et al. Protein arginine methyltransferase 6 enhances polyglutamine-expanded androgen receptor function and toxicity in spinal and bulbar muscular atrophy. Neuron. 2015;85(1):88-100. doi:10.1016/j.neuron.2014.12.031