Dutasteride Reduces Viral Shedding, Inflammatory Responses and Time-to-Remission in COVID-19: Biochemical Findings of a Randomized Double-Blind Placebo Controlled Interventional Trial (DUTA AndroCoV-Trial - Biochemical).

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Research Article

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Abstract

Importance: SARS-CoV-2 cell entry and infectivity is indirectly dependent on androgenic status and phenotype through the regulation of transmembrane protease serine 2 (TMPRSS2), an androgen-mediated proteolytic enzyme that facilitates SARS-CoV-2 entry. Males, particularly those affected by androgenetic alopecia (AGA) are overrepresented in severe COVID-19, while the use of 5-alpha-reductase inhibitors (5ARis), an antiandrogenic drug class, may reduce COVID-19 severity.

Objective: Our objective was to determine if dutasteride, a wide and potent 5ARi, would bring biochemical and virological benefits in early COVID-19.

Design, Setting, and Participants: A double-blinded, randomized, prospective, investigational study of dutasteride for the treatment of COVID-19, as add-on therapy to the local standard of care, for mild or moderate, non-hospitalized subjects confirmed for SARS-CoV-2 (The Duta AndroCoV Trial).

Interventions: Dutasteride 0.5mg/day or placebo for 30 days or until full COVID-19 remission. Nitazoxanide was given 500mg twice a day for six days and azithromycin was given 500mg/day for five days for all subjects.

Main Outcome(s) and Measure(s): Remission times for fatigue, ageusia, anosmia, and overall disease, oxygen saturation (%), real-time polymerase chain reaction (rtPCR-SARS-CoV-2), ultrasensitive C-reactive protein (usCRP), D-dimer, lactate, dehydrogenase lactate (DHL), erythrocyte sedimentation rate (ESR), ultrasensitive troponin and ferritin.

Results: Compared to placebo group (n=44) with similar baseline characteristics, dutasteride (n=43) presented reduced fatigue, anosmia and overall disease duration (46.6%, 49.6% and 43.2% lower duration, respectively; p<.0001 for all), and in Day 7 presented higher rates of virologic cure (64.3% versus 11.8% cure; p=.0094), , increased recovery rate (84.7% versus 57.5%; p=.03), higher mean [SD] oxygen saturation (97.0% [1.4%] versus 95.7% [2.0%]; p=.02), lower median [IQR] usCRP (0.34mg/L [0.23mg/L -0.66mg/L] versus 1.47mg/L [0.70mg/L-3.37mg/L]; p<.0001), lower median [IQR] lactate (2.01mmol/L [1.12mmol/L-2.43mmol/L] versus 2.66mmol/L [2.05mmol/L-3.55mmol/L]; p=.0049), lower median [IQR] ESR (5.0mm/1h [3.0mm/1h-11.0mm/1h] versus 14.0mm/1h [7.25mm/1h-18.5mm/1h]; p=.0007), lower median [IQR] LDH (165U/L [144U/L -198U/L] versus 210U/L [179U/L-249U/L]; p=.0013 and lower median [IQR] troponin levels (0.005ng/mL [0.003ng/mL-0.009ng/mL] versus 0.007ng/mL [0.006ng/mL-0.010ng/mL]; p=.048).

Conclusions and Relevance: These findings suggest that dutasteride reduces clinical and virologic disease duration and inflammatory markers in males with mild-to-moderate, early-stage COVID-19, and should be considered as a therapeutic option in the current context of the COVID-19 pandemic.

Trial Registration: NCT04446429

Introduction

COVID-19 pandemic disproportionately affects men, since men are more likely to be infected, more likely to have severe disease, and have a greater case fatality rate compared to women.[1,2], which is not fully explained by sex disparities in terms of lifestyle and comorbidities. We have further detected that androgen-mediated phenotype of androgenetic alopecia (AGA) in males and hyperandrogenism states in females are linked to COVID-19 disease severity.[3-5], while the use of antiandrogens was correlated with lower severity[6-8]

SARS-CoV-2 entry into cells is dependent on a modification of a viral spike protein by the transmembrane protease, serine 2 (TMPRSS2) expressed on the surface of human cells, which the only known modulators androgens, through the androgen response element located in the 5' promoter region[9,10]. It is plausible to hypothesize that SARS-CoV-2 viral infectivity is regulated by androgens, as explained in several communications that we have published suggesting that the male bias in COVID-19 disease severity may be linked to androgens, and reinforced by the disease patterns according to the androgenic phenotypes in both males and females[3-5;11-13]. Accordingly, the reduction of the TMPRSS2 expression by blocking the androgen receptor would decrease SARS-CoV-2 entry into human cells[13-17], which is corroborated by studies showing protection from more severe states related to COVID-19 with the use of antiandrogens[6-8]. In addition, variation in the androgen receptor (AR) gene may predict COVID-19 severity[13,18].

Taken together, there is sufficient substantiation to hypothesize that the use of drugs that reduce androgen receptor function may be a promising target against COVID-19.

5-alpha-reductase inhibitors (5ARis) are commonly prescribed antiandrogens for androgenetic alopecia and benign prostatic hyperplasia. Their mechanism of action is the blockage of the conversion of testosterone to dihyrotestosterone (DHT), a five-times more potent androgen.[11] 5ARis are inexpensive and have relatively low incidence of adverse side effects. Because of our previous reports on the protective role of dutasteride when used in a chronic, regular basis, associated with the increasing evidence and evident mechanistical plausibility of the promising roles of antiandrogens as protective agents, we conducted a double blind, placebo-control randomized clinical trial (RCT), the Duta AndroCoV Trial, aimed to determine whether the use of dutasteride during early COVID-19 would be beneficial in terms of disease outcomes.

In addition to the clinical outcomes evaluated in all participants enrolled in the study, two thirds of the subjects of the Duta AndroCoV Trial also underwent biochemical analysis, in a 2:1 randomized distribution, aiming to detect differences in inflammatory and virological responses between dutasteride and placebo. The objective of the present manuscript is to describe the major findings of the biochemical analysis of the Duta AndroCoV Trial.

Methods

Study Design

Potential subjects were recruited to a double-blinded, randomized, prospective, investigational study of anti-androgen treatment of COVID-19 (NCT04446429) through social media and also a mailing list containing 10,900 men from a Brasilia-based Brazilian health care system registry and were also referred from external outpatient settings. For the present study, subjects presented confirmed COVID-19 infection with mild symptoms and did not require admission to a hospital at the time of their first visit. The study is registered (clinicaltrial.gov) and conducted with the approval of the Brazilian National Ethics Committee (Approval number 4.173.074; process number (CAAE) 34110420.2.0000.0008; Comitê de Ética em Pesquisa (CEP), Comitê Nacional de Ética em Pesquisa (CONEP), Ministry of Health (Ministério da Saúde (MS)) (CEP/CONEP/MS). All patients admitted to the study signed informed consent.

Baseline characteristics, presence of comorbidities, use of medications, clinical characterization of COVID-19, test results and disease outcomes were recorded by the principal investigator and managed by the study director.

Study Population

Screening of subjects suspected for COVID-19 was conducted by the principal investigator centralized at the major site of the research (Corpometria Institute Brasilia, Brazil), at which nasopharyngeal swabs were collected by trained medical personal. SARS-CoV-2 status was laboratory confirmed by real-time reverse transcription polymerase reaction (rtPCR-SARS-CoV-2) testing (Automatized Platform, Roche, USA) following the Cobas SARS-CoV-2 rtPCR kit test protocol. Subjects already confirmed for COVID-19 using rtPCR-SARS-CoV-2 and that fulfilled criteria for the study were enrolled directly.

Additional inclusion criteria included: 1. Above 18 years old; 2. Confirmation of COVID-19 infection for less than seven days; 3. Absence of specific treatments for COVID-19 for periods longer than 72 hours; 4. Absence of contraindications to any of the molecules tested in the present RCT; 5. Oxygen saturation ($SatO_2$) above 92% at the time of the first visit; and 6. No signs of complications related to COVID-19 and secondary bacterial infections.

Procedures

Patients were randomized for either dutasteride or placebo groups. Dutasteride was given 0.5mg/day for 30 days or until full COVID-19 remission. For all subjects, nitazoxanide was given 500mg twice a day after meal for six days and azithromycin was given 500mg/day one hour before meal for five days, as per one of the standardized therapies suggested by the local Ministry of Health.

A 2:1 randomization of subjects enrolled in the DUTA Andro-CoV Trial (n = 130), underwent biochemical exams in at least two times between Day 0, Day 7 and Day 14, and were selected for the present analysis. Clinical outcomes were also adjusted for the subset of patients that were selected for the present study.

Study Outcomes

Endpoints for the study were remission times for fatigue and loss of taste or smell (ageusia or anosmia), time to overall COVID-19 remission including and not including anosmia and ageusia. Disease severity was rated at Day 0 (start of treatment), Day 1, Day 2, Day 3, Day 7, Day 14, Day 21, Day 30, and Day 60, according to a their health on a scale from 0 to 100, 100 being completely heathy and 0 being their worst day of COVID-19. Percentage of patients completely free of clinical symptoms of COVID-19 was determined at Day 7. Oxygen saturation (%) and heart rate (bpm) were also measured in Day 0, Day 1, Day 2, Day 3 and Day 7.

Biochemical parameters included for rtPCR-SARS-CoV-2 in Day 7, and ultrasensitive C-reactive protein (usCRP) (mg/L) (latex-intensified immunoturbidimetry), D-dimer (ng/mL) (immunologic assay), lactate (mmol/L) (enzymatic assay), dehydrogenase lactate (DHL) (U/L) (lactate NAD), erythrocyte sedimentation rate (ESR) (mm/1h) (capillary photometry), ultrassensitive troponin (ng/mL) (electric chemiluminescence - ECLIA) and ferritin (ng/mL) (chemiluminescence – CLIA), in pre-COVID (if available), Day 0, Day 7, Day 14, D Day 7-0, D Day 14-0, and percentage of different levels of reductions between Days 0 and 7 and between Days 0 and 14.

Additional records included neutrophils (*1000/mm^3), lymphocytes (*1000/mm^3), eosinophils (*1000/mm^3) and monocytes (*1000/mm^3) (Flow cytometry with fluorescent protein – XN10-Sysmex), and neutrophil-to-lymphocyte ratio was then calculated.

Sample Size Calculations

Based on the assumptions that sample size should be estimated for the chi-squared test to detect the difference in proportions at p=0.05, that the number of subjects enrolled in Arm A is equal to the number of subjects in Arm B i.e., 1:1 enrollment ration, and that 95% of subjects will complete the study, we calculated[3] that at a minimum we needed to recruit a minimum number of 127 subjects.

Statistical Analysis

Medical history, concomitant medications and lifestyle characteristics of COVID-19 patients were tabulated based for each group: age, BMI, hypertension, myocardial infarction, stroke, heart failure, lipid disorders, diabeties, pre-diabeties, obesity, asthma, COPD, cancer, benign prostatic hyperplasia, prostate cancer, chronic renal disease, liver fibrosis/cirrhosis, clinical depression, anxiety, ADHD, insomnia, hypogonadism, hypothyroidism, and autoimmune disorders as well as indicated medications. Average and standard deviation (SD) levels for clinical parameters and median and 95% confidence interval (95% CI) for biochemical parameters were calculated. Non-parametric statistical tools were employed to determine statistical significance, which was set at $p<0.05$. XLSTAT version 2020.3.1.1008 (Addinsoft, Inc.) was used to perform all statistical analysis.

Results

Of the 130 SARS-CoV-2 males included in the Duta AndroCoV trial, 87 underwent a sequence of biochemical analyses and were included in the present study. Of these, 44 men were included in the investigational arm and 43 men in the placebo group (**Figure 1**). Average interval between first symptoms and beginning of treatment was 4.3 days for both groups (p = n/s). Baseline characteristics, prevalence of comorbidities and use of medications in the two study groups are displayed in **Table 1** and were similar for all parameters.

Table 1. Characteristics of the study populations.

	Placebo (n = 43)	Dutasteride (n = 44)	p-value
	AVG (STD)	AVG (STD)	
AGE (y/o)	43.8 (14.1)	40 (10.8)	n/s
BMI (Kg/m2)	26.1 (2.2)	26.0 (3.2)	n/s
Time-to-treat	4.4 (1.4)	4.3 (1.4)	n/s
	N (%)	N (%)	
Obesity	4 (9.3%)	5 (11.4%)	n/s
Hypertension	12 (27.9%)	7 (15.9%)	n/s
Mi	0	0	n/a
Stroke	0	0	n/a
Hf	0	1 (2.3%)	n/a
Lipid disorders	9 (20.9%)	9 (20.4%)	n/s
Other cardiac dysfunctions	0	1 (2.3%)	n/a
Diabetes	4 (9.3%)	4 (9.1%)	n/s
Pré-dm2	8 (18.6%)	5 (11.4%)	n/s
Dysglycemia	12 (28.0%)	9 (20.6%)	n/s
Asthma	1 (2.3%)	0	n/a
COPD	0	0	n/a
Chronic renal dis.	0	0	n/a
Liver fibrosis/chirrosis	1 (2.3%)	0	n/a
Clinical depression	2 (4.6%)	3 (6.8%)	n/s
Anxiety	6 (13.9%)	8 (18.2%)	n/s
ADHD	5 (11.6%)	3 (6.8%)	n/s
Insomnia	4 (9.3%)	1 (2.3%)	n/s
Hypothyroidism	1 (2.3%)	4 (9.1%)	n/a
Autoimmune disorders	1 (2.3%)	1 (2.3%)	n/a
Cancer	0	0	n/a

Hypogonadism	8 (18.6%)	8 (18.2%)	n/a
BPH	2 (4.6%)	2 (4.5%)	n/s
DRUGS	N (%)	N (%)	
Beta-blocker	2 (4.6%)	1 (2.3%)	n/a
ECAi	1 (2.3%)	0	n/a
ARB	11 (25.6%)	7 (15.9%)	n/s
Loop diur.	1 (2.3%)	0	n/a
Thiazide diuretics	4 (9.3%)	1 (2.3%)	n/a
CCB	2 (4.6%)	2 (4.5%)	n/s
Statins	8 (18.6%)	8 (18.2%)	n/s
Others	0	0	n/a
Aspirin	0	1 (2.3%)	n/a
Clopidogrel	0	0	n/a
Warfarin	0	0	n/a
Xa factor inhibitors	0	0	n/a
Direct thrombin inhibitors	0	0	n/a
Heparins	0	0	n/a
Metformin	10 (23.3%)	6 (13.6%)	n/s
GLP1R analogue	2 (4.6%)	3 (6.8%)	n/a
SGLT2 inhibitors	7 (16.3%)	3 (6.8%)	n/s
DPP4 inhibitors	2 (4.6%)	1 (2.3%)	n/a
Sylfonylureas	0	0	n/a
Glitazone	0	0	n/a
Acarbose	0	0	n/a
Insulin	0	0	n/a
Orlistat	2 (4.6%)	2 (4.5%)	n/a
Levothyroxine	1 (2.3%)	4 (9.1%)	n/a
Liothyronine	1 (2.3%)	0	n/a
Testosterone	7 (16.3%)	6 (13.6%)	n/s

Aromatase inhibitors or SERMs	2 (4.6%)	2 (4.5%)	n/s
Hipnotics	3 (7.0%)	1 (2.3%)	n/a
Selective serotonin reuptaker inhibitors (SSRI)	3 (7.0%)	6 (13.6%)	n/s
Other antidepressants and humor stabilizers	4 (9.3%)	4 (9.1%)	n/s
Benzodiazepines	1 (2.3%)	0	n/a
Atypical antipsychotics	3 (7.0%)	2 (4.5%)	n/a
CNS stimulants	5 (11.6%)	5 (11.4%)	n/s
Alpha-1 adren. Blockers	2 (4.6%)	2 (4.5%)	n/s
Gnrh analogues and inh., NSAA, others	0	0	n/a
Erectile dysfunction	2 (4.6%)	1 (2.3%)	n/a
Omega-3	3 (7.0%)	1 (2.3%)	n/a
Vitamin D	13 (30.2%)	15 (34.1%)	n/s
Zinc	6 (13.9%)	7 (15.9%)	n/s
Biotin	1 (2.3%)	0	n/a
Vitamin C	8 (18.6%)	7 (15.9%)	n/s
Multivitamin	1 (2.3%)	2 (4.5%)	n/a
BCG	43 (100%)	44 (100%)	n/s
Influeza (in 2020)	14 (32.6%)	14 (31.8%)	n/s
Pneumococcal (since 2017)	3 (7.0%)	5 (11.4%)	n/s
Current smoking	2 (4.6%)	2 (4.5%)	n/a
Regular physical activity	28 (65.1%)	32 (72.7%)	n/s
ADDITIONAL COVID TREATMENTS			
Ivermectin	6 (13.9%)	6 (13.6%)	n/s
Hydroxychlorouquine	4 (9.3%)	3 (6.8%)	n/s
Xa factor inhibitors	8 (18.6%)	3 (6.8%)	n/s
Enoxaparin	3 (7.0%)	3 (6.8%)	n/s
Glucocorticoids	6 (13.9%)	7 (15.9%)	n/s
Vitamin c	4 (9.3%)	4 (9.1%)	n/s
Zinc	3 (7.0%)	3 (6.8%)	n/s

Vitamin d	1 (2.3%)	2 (4.5%)	n/a
Colchicine	0	0	n/a
Bromexhine	0	0	n/a
N-acetylcysteine	0	0	n/a

None of the parameters was statistically significant between groups.

Congestive heart failure = CHF; type 2 diabetes mellitus = T2DM, chronic obstructive pulmonary disorder = COPD, chronic kidney disease = CKD, attention deficit hyperactivity disorder = ADHD, benign prostate hyperplasia = BPH, angiotensin converting enzyme inhibitors = ACEi; angiotensin-2 receptor blockers = ARB; calcium channel blocker = CCB; glucagon-like peptide-1 receptor analogues = GLP1Ra; sodium-glucose cotransporter-2 inhibitors = SGLT2i; dipeptidyl-peptidase 4 inhibitors = DPP4i; progesterone = P; estradiol = E; gonadotropin release hormone = GnRH; selective estrogen receptor modulators = SERM; non-steroidal antiandrogen = NSAA; selective serotonin reuptake inhibitors = SSRI; central nervous system = CNS; Bacillus Calmette-Guérin = BCG.

Table 2 reports the average times to remission for major clinical symptoms and overall time to remission, patient reported outcome rating disease severity in Days 1, 2, 3 and 7, and oxygen saturation in Days 0, 7 and 14, in men taking 5ARis versus men not taking a 5ARi.

The average (± standard deviation) remission time for fatigue was 5.5 (±3.2) days in the dutasteride group versus 10.3 (±8.4) days in the placebo group ($p < 0.001$), or 46.6% reduction. The average remission time for loss of taste or smell was 5.6 (±4.0) days in the dutasteride group versus 11.1 (±6.6) days in the placebo group ($p < 0.001$), or 49.6% reduction. Time to full remission was 9.2 (±4.3) days in the dutasteride group versus 16.3 (±8.3) days in the placebo group (43.2% reduction; $p < 0.001$). When anosmia and ageusia are excluded from the analysis, the average time to full remission was 7.0 (±2.9) days in the dutasteride group versus 11.7 (±7.7 (43.6% reduction; $p < 0.001$).

Level of recovery was significantly improved in the dutasteride group compared to the placebo group for Days 1, 2, 3 and 7 ($p < 0.0001$ for all days). The percentage of

patients still affected at Day 7 was 15.9% in the dutasteride group and 42.5% in the placebo group.

Mean oxygen saturation was statistically higher in the dutasteride group for Days 7 ($p = 0.02$) and 14 ($p = 0.0012$), as well as level of oxygen saturation increase between Days 0 and 7 ($p = 0.047$).

Table 2. Clinical outcomes.

Mean ± SD	Placebo (n=66)	Dutasteride (n=64)	*p-value*
Time-to-remission *(Mean ± SD)*			
Fatigue	10.3 (±8.4)	5.5 (±3.2)	**< 0.001**
Loss of Taste or Smell (Ageusia or Anosmia)	11.1 (±6.6)	5.6 (±4.0)	**< 0.001**
Remission Minus Taste or Smell Loss	11.7 (±7.7)	7.0 (±2.9)	**< 0.001**
Overall symptoms	16.3 (±8.3)	9.2 (±4.3)	**< 0.001**
Clinical recovery *(Mean ± SD)*			
% fully clinically recovered at Day 7	57.5%	84.1%	**0.03**
% of clinical recovery at Day 1	34.2 (±21.4)	60.4 (±24.2)	**< 0.0001**
% of clinical recovery at Day 2	52.9 (±21.3)	78.5 (±17.8)	**< 0.0001**
% of clinical recovery at Day 3	66.8 (±20.7)	89.2 (±12.3)	**< 0.0001**
% of clinical recovery at Day 7	82.9 (±15.0)	97.4 (±5.7)	**< 0.0001**
Oxygen saturation (%) *(Mean ± SD)*			
Day 0	95.4 ± 1.4	96.0 ± 1.5	n/s (0.21)
Day 7	95.7 ± 2.0	97.0 ± 1.4	**0.02**
Day 14	96.2 ± 1.4	97.5 ± 1.2	**0.0012**
D Day 7-0	+0.3	+1.3	**0.047**
D Day 14-0	+0.9	+1.3	n/s (0.25)

Both groups used nitazoxanide plus azithromycin.

n/s = non significant; n/a = non applicable; STD = standard deviation

Table 3 displays biochemical parameters in the dutasteride and placebo groups, including the percentage of subjects with negative rtPCR SARS-CoV-2 in Days 0, 7 and 14, and levels of usCRP, lactate, ESR, LDH, ultrasensitive troponin, D-dimer and ferritin levels in Days 0, 7, 14, changes between Days 0 and 7 and between Days 0 and 14, and percentage of men with specific goals for each parameter between Days 0 and 7 and between Days 0 and 14.

In Day 7, 64.7% and 11.8% of men from the dutasteride and placebo group were free from SARS-CoV-2 virus or fragments, respectively (increase of 444.9% in the percentage of subjects cured in Day 7; $p = 0.0094$). In Day 14, 88.3% of the dutasteride group and 54.2% of the placebo group yielded non-detectable SARS-CoV-2.

In Day 7, median usCRP was 0.34 in the dutasteride group and 1.47 in the placebo group ($p < 0.0001$). In Day 14, median usCRP was 0.36 in the dutasteride group and 0.39 in the placebo group ($p = 0.0026$). Compared baseline levels, usCRP was reduced in Day 7 in 83.4% of men from the dutasteride and 64.7% of men from the placebo group, and in 93.7% and 73.3% of men from dutasteride and placebo group, respectively, in Day 14.

In Days 7 and 14 men from the dutasteride group had lower lactate levels than placebo group ($p = 0.0049$ and $p = 0.014$ for Days 7 and 14, respectively, and lack of lactate increase above 0.5 mmol/L between Days 0 and 7 was observed in 60% of subjects of the dutasteride group and 19.2% of subjects of the placebo group ($p = 0.007$).

Median ESR was 5.0 mm/1h in the dutateride group and 14.0 mm/1h in the placebo group in Day 7 ($p = 0.0007$), and 4.0 mm/1h in the dutasteride group and 11.5 mm/1h in the placebo group ($p < 0.0001$) in Day 14. Between Days 0 and 7, ESR reduced more than 10 mm/1h in 41.4% of men from the dutasteride group and 16.1% of those from the placebo

group. Between Days 0 and 7, ESR reduced more than 10 mm/1h in 59.4% of men from the dutasteride group and 30.0% of those from the placebo group

LDH levels were statistically lower in the dutasteride group than placebo group in Day 7 ($p = 0.0013$) and Day 14 ($p = 0.0004$).

In Day 7, ultrasensitive troponin levels were significantly lower in the dutasteride group (median = 0.005 ng/mL) than placebo group (median = 0.007 ng/mL) ($p = 0.048$). In Day 7, ultrasensitive troponin levels reduced more than 0.003 ng/mL in 80% of participants in the dutasteride group and 52.8% in the placebo group. In Day 7, ultrasensitive troponin levels reduced more than 0.003 ng/mL in 66.7% of participants in the dutasteride group and 33.3% in the placebo group.

The only parameter that disclosed significant differences in D-dimer levels was in Day 14, with median D-dimer of 220 ng/mL in the dutasteride group and 305 ng/mL in the placebo group ($p = 0.019$). Ferritin levels and changes were similar between dutasteride and placebo group for all times and intervals.

No severe adverse effects, hospitalizations, mechanical ventilation or deaths were reported. **Figure 2** summarizes the main clinical and biochemical findings of the present study.

Table 3. Biochemical results.

	Placebo (n=66)	Dutasteride (n=64)	*p-value*
rtPCR SARS-CoV-2 remission *(CT > 40 cycles) (%)*			
Day 0	0%	0%	n/s (1.00)
Day 7	11.8%	64.3%	**0.0094**
Day 14	54.2%	88.3%	**0.036**
usCRP (mg/L) *Median (95% CI)*			
Pre-COVID	0.18 (0.08-0.41)	0.38 (0.10-0.56)	n/s (.45)
Day 0	1.44 (0.72-2.54)	1.22 (0.83-2.26)	n/s (.42)
Day 7	1.47 (0.70-3.37)	0.34 (0.23-0.66)	**< .0001**
Day 14	0.39 (0.21-2.32)	0.36 (0.08-0.34)	**.0026**
usCRP decrease D Day 7-0	64.7%	83.9%	n/a
usCRP decrease D Day 14-0	73.3%	93.7%	n/a

Lactate (mmol/L) *Median (95% CI)*			
Pre-COVID	0.93 (0.82-1.18)	0.88 (0.72-1.23)	n/s (.58)
Day 0	1.72 (1.35-2.11)	1.51 (1.16-2.01)	n/s (.27)
Day 7	2.66 (2.05-3.55)	2.01 (1.12-2.43)	**.0049**
Day 14	1.92 (1.38-2.89)	1.48 (1.22-1.89)	**.014**
D Day 7-0	+0.97 (+0.57 - +1.69)	+0.28 (+0.02 - +0.85)	**.025**
ate increase < 0.5mmol/L DDay(7-0)	19.2%	60%	n/a
D Day 14-0	+0.22 (-0.64 - +0.79)	-0.19 (-1.15 - +0.27)	n/s (.24)
te increase < 0.5mmol/L DDay(14-0)	67.8%	83.3%	n/a
ESR (mm/1h) *Median (IQT)*	**Placebo** **(n = 43)**	**Dutasteride** **(n = 44)**	***p-value***
Pre-COVID	4.5	4.0 (2.0-7.0)	n/s (.76)

	(2.25-8.0)		
Day 0	13.5 (7.0-22.5)	13.0 (6.5-22.0)	n/s (.69)
Day 7	14.0 (7.25-18.5)	5.0 (3.0-11.0)	**.0007**
Day 14	11.5 (6.5-18.0)	4.0 (3.0-5.0)	**< .0001**
D Day 7-0	-4.0 (-6.75 - +4.5)	-8.0 (-13.0 – -1.0)	**.017**
₹R decrease > 10mm/1h DDay(7-0)	16.1%	41.4%	n/a
D Day 14-0	-4.5 (-11.0 - +4.25)	-11.5 (-19.25 - -2.75)	**.003**
R decrease > 10mm/1h DDay(14-0)	30%	59.4%	n/a
LDH (U/L) *Median (95% CI)*			
Pre-COVID	175 (158-211)	183 (156-199)	n/s (.76)
Day 0	207 (175-234)	200 (172-222)	n/s (.69)
Day 7	210 (179-249)	165 (144-198)	

			.0013
Day 14	177 (154-202)	147 (135-160)	
			.0004
D Day 7-0	+4 (-32.5 - +40.25)	-21 (-53 - +18)	
			.087
	26.5%	50.0%	
duction > 30 U/L between Days 0 and 7			
			n/a
D Day 14-0			
	-26 (-68 - -10)	-35 (-76 - -15)	n/s (.31)
duction > 30 U/L between Days 0 and 7			
	46.4%	64.3%	n/a
Ultrassensitive troponin (ng/mL) *Median (95% CI)*			
Pre-COVID	n/a	n/a	n/a
Day 0	0.008 (0.005-0.012)	0.010 (0.008-0.015)	n/s (.23)
Day 7	0.007 (0.006-0.010)	0.005 (0.003-0.009)	**.048**
	0.005	0.004	

	(0.004-0.008)	(0.003-0.005)	
Day 14			n/s (.14)
D Day 7-0	-0.003 (-0.005 – 0)	-0.004 (-0.007 – -0.003)	.094
Troponin decrease > 0.003ng/mL DDay(7-0)	**52.8%**	**80%**	n/a
D Day 14-0	-0.003 (-0.007 – -0.001)	-0.007 (-0.011 – -0.004)	.16
Min decrease > 0.005ng/mL DDay(14-0)	**33.3%**	**66.7%**	n/a
D-dimer (ng/mL) *Median (95% CI)*			
Pre-COVID	343 (215-427)	366 (255-438)	n/s (1.00)
Day 0	404 (289-575)	406 (266-584)	n/s (.77)
Day 7	310 (220-495)	189 (242-373)	n/s (.29)
Day 14	305 (216-420)	220 (200-306)	**.019**

D Day 7-0	-80 (-182 - +52)	-104 (-190 - -35)	n/s (.30)
D-dimer decrease D Day 7-0	63.6%	84.2%	n/a
D Day 14-0	-70 (-241 - +31)	-137 (-325 - -16)	n/s (.20)
ıer decrease > 100ng/mL D Day 14-0	42.8%	61.6%	n/a
Ferritin (ng/mL)			
Median (95% CI)			
Pre-COVID	173 (133-241)	190 (113-310)	n/s (.61)
Day 0	389 (295-588)	471 (279-584)	n/s (.85)
Day 7	321 (248-476)	310 (198-476)	n/s (.41)
Day 14	240 (186-377)	241 (180-352)	n/s (.66)
D Day 7-0	-83 (-139 - -52)	-68 (-214 - -9)	n/s (.99)
D Day 14-0	-128 (-262 - -72)	-135 (-289 - -55)	n/s (.82)

Both groups used nitazoxanide plus azithromycin.

IQT = interquartile n/s = non significant; n/a = non applicable; rtPCR = real time polymerase chain reaction; usCRP: ultrasensitive C-reactive protein; ESR = erythrocyte sedmentation rate; LDH = lactate dehydrogenase

Discussion

The detection of effective methods against COVID-19 is critical while vaccines are not widely available. To date, poor quality studies have been conducted in actual early COVID-19, while more than 98% of larger RCTs were performed in further stages, even when alleging that these studies were performed during early-stage COVID-19. These limitations preclude us from conclusive findings on the efficacy of any drug regimen.

Results of parameters evaluated in both overall males included in the Duta AndroCoV Trial and the present analysis of a subset of patients were similar. Baseline characteristics and comorbidities prevalence were similar between groups, which reduces the chances of differences between responses to COVID-19 be secondary to external factors.

Similarly to what was observed in the RCT that included all subjects, the subset population included in the present analysis demonstrated reductions between 40% and 50% in symptoms and overall time to remission. Differences in the recovery process could be detected as early as in Day 1, showing an approximate two times better recovery than controls. In Day 7, three times less men from the dutasteride group remained symptomatic than men from the placebo group. Accordingly, oxygen saturation demonstrated a faster recovery speed among subjects from the dutasteride group.

Virological cure for COVID-19 can be determined by a negative rtPCR SARS-CoV-2, since while detected virus may not necessarily reflect the presence of viable and alive virus, a negative result after confirmation of COVID-19 has almost 100% certainty for cure. More than five times more males from the dutasteride group compared to those from the placebo group were cured from COVID-19 in Day 7, and four times more males from the placebo group maintained positive rtPCR than males from the dutasteride group in Day 14. The substantial differences provide overwhelming evidence for the antiviral activity of dutasteride during early COVID-19.

Multiple reductions in biochemical parameters related to inflammatory responses to SARS-CoV-2, including reductions in usCRP, lactate, ESR and LDH levels in the dutasteride group compared to the placebo group, provide additional evidence for the protective role of dutasteride in COVID-19. Although inflammation was not overtly detected in none of the groups, the blockage of the progression to inflammatory states, even when mild, may have contributed to the shortened disease duration. Indeed, biochemical findings meet clinical improvements presented by the dutasteride group, reinforcing its potential protective actions against COVID-19.

Overall, differences were more substantial in Day 7 than Day 14, which allow us hypothesize that the most important impact of dutasteride is to increase the speed of COVID-19 recovery.

While androgens are both circulating and produced in tissue, elevated tissue DHT, implicated in AGA and benign prostatic hyperplasia, and prostate cancer, as well as AR sensitivity, may be better predictors of COVID-19 severity than circulating androgen levels.

Changes in serum testosterone and DHT in response to dutasteride occur as early as one to two days after dutasteride initiation, shorter than the period comprised by the present RCT[19]. In addition, in-cell and within tissue concentration of these hormones occurs earlier than changes observed in the sera[20], eventually leading to a rapid reduction of TMPRSS-2 expression that may help explain the efficacy of dutasteride observed through our double blind, placebo-control RCT.

Findings of the present analysis are in accordance with the observational study (The Pre-AndroCoV Trial), which demonstrated that the addition of dutasteride or spironolactone enhanced the response against COVID-19 when treated with hydroxychloroquine with or without ivermectin or nitazoxanide, combined with azithromycin[21].

Whether dutasteride is effective against COVID-19 when used alone is uncertain although likely. When used chronically, evidence suggests a protective role of dutasteride in reducing COVID-19 triggered complications. The combination of different drugs working synergistically follows the rationale of the complex SARS-CoV-2 pathogenicity and infectivity mechanisms of action.

The increased speed of recovery with the use of dutasteride may be particularly useful for patients between Day 4 and Day 7 of symptoms, during the late period of the early stage of COVID-19, when approaches should induce sufficiently fast response in order to avoid progression to an inflammatory state and consequent acute lung injury.

Results presented herein were detected even in the challenging heterogeneity of COVID-19 symptoms and disease course, which precludes from normally distributed results and hampers the detection of effective therapies, unless when differences between the drug and placebo or control are substantial.

Here we demonstrated in a randomized, double-blinded, placebo controlled interventional study that men treated with dutasteride, a comprehensive and potent 5ARi, reduces viral duration, clinical symptoms, disease course, and inflammatory responses induced by COVID-19 in an outpatient setting when combined to nitazoxanide and azithromycin, compared to nitazoxanide and azithromycin alone. Whether results obtained with dutasteride can be observed in other 5ARis is uncertain. Stronger antiandrogens, such as proxalutamide, a novel non-steroidal anti-androgen (NSAA), may provide further benefits in the treatment of COVID-19, and is being currently investigated in a double-blind placebo-control RCT by our group (NCT04446429).

Conclusion

Dutasteride reduced COVID-19 viral shedding, course duration and inflammation when combined with nitazoxanide and azithromycin, when compared to nitazoxanide and azithromycin alone, in a randomized, double-blinded, placebo controlled interventional study in males.

Declarations

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Conflict of interest statement

Authors declare no conflict of interest with any of the pharmacological interventions proposed by the present study.

Data availability

Full raw data is available under request to Flavio.cadegiani@unifesp.br, the principal investigator of the study.

Ethics approval statement

The present study was approved by the Institutional Review Board (IRB) of the Ethics Committee of the National Board of Ethics Committee of the Ministry of Health, Brazil (CEP/CONEP: Parecer 4.173.074 / CAAE: 34110420.2.0000.0008), and is registered at ClinicalTrials.gov (Identifier: NCT04446429. Available at clinicaltrials.gov (https://clinicaltrials.gov/ct2/show/NCT04446429?term=NCT04446429&draw=2&rank=1).

Patient consent statement

Patients included for the present analysis provided a written consent exactly as approved by the Institutional Review Board (IRB) of the Ethics Committee of the National Board of Ethics Committee of the Ministry of Health, Brazil (CEP/CONEP: Parecer 4.173.074 / CAAE: 34110420.2.0000.0008), alongside with the currently ongoing randomized clinical trial (RCT) registered at ClinicalTrials.gov (Identifier:

NCT04446429. Available at clinicaltrials.gov (https://clinicaltrials.gov/ct2/show/NCT04446429?term=NCT04446429&draw=2&rank=1).

Authorship

Authors FAC, AG, CGW and JM contributed equally for the present study. FAC, AG, CGW and JM designed the study. FAC enrolled, provided medical assistance to all patients and compiled all data, while FAC, AG, CGW and JM contributed for the analysis of the data and development of the manuscript in its current format.

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