STATISTICAL ANALYSIS PLAN

Study: Efficacy and Safety of Edoxaban and or Colchicine for patients with SARS-CoV-2 infection managed in the out of hospital setting (COVID 19)

Short Title: CorONa VIrus edoxabaN ColchicinE (CONVINCE)

Date: 09.11.2020
Version: 1 (Protocol version: V3/04.11.2020)

Version History		
Version Number: 1	Date: 09.11.2020	Total number of pages: 15

AUTHOR

Dr. Greta Carrara
Statistic
Advice Pharma Group

APPROVAL SIGNATURES

The signatories have read the statistical analysis plan of the study titled "Efficacy and Safety of Edoxaban and or Colchicine for patients with SARS-CoV-2 infection managed in the out of hospital setting."- Version 1, dated 09 November 2020 - carefully and agree to adhere to its provisions.

Author	**Signature**	**Date**
Dr. Greta Carrara Statistician Advice Pharma Group		12/11/2020

Sponsor	**Signature**	**Date**
Prof. Stephan Windecker Insel Gruppe AG,Inselspital, Cardiology Bern, Switzerland		13/11/20

Coordinating Principal Investigator	**Signature**	**Date**
Prof. Marco Valgimigli Cardiocentro Ticino Lugano, Switzerland		12/11/2020

Coordinating Principal Investigator	**Signature**	**Date**
Prof. Pascal Vranckx Hasselt, Belgium	pascal vranckx	13/11/2020

Coordinating Principal Investigator	**Signature**	**Date**
Dr. Nuccia Morici ASST Grande Ospedale Metropolitano Niguarda Milan, Italy		12/11/2020

TABLE OF CONTENTS

LIST OF ABBREVIATIONS

AE	Adverse Event
BARC	Bleeding Academic Research Consortium
CI	Confidence Interval
CNS	Central nervous system
COVID-19	Coronavirus disease of 2019
CRP	C-reactive protein
DVT	Deep vein thrombosis
ECG/EKG	Electrocardiography
(e)CRF	(electronic)Case Report Form
FAS	Full analysis
HS	High sensitive
ISTH	International Society of Thrombosis and Hemostasis
MedDRA	Medical Dictionary for Regulatory Activities
mITT	Modified Intention-to-treat
MOF	Multiorgan failure
MVTE	Major vascular thrombotic events
NSAIDs	Nonsteroidal anti-inflammatory drugs
PE	Pulmonary embolism
PP	Per-protocol
PT	Preferred Term
RT PCR	Reverse transcriptase-polymerase chain reaction
SAE	Serious Adverse Event
SARS-CoV-2	Coronavirus responsible for COVID-19
SOC	Primary System Organ Class
VTE	Venous thromboembolism

1. INTRODUCTION

First appearing in Wuhan, China, the coronavirus disease of 2019 (COVID-19) is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Given the rapid spread of this virus with consequences on an international scale, COVID-19 was declared a pandemic by the World Health Organization on March 11th 2020.

There is emerging evidence that patients with SARS-CoV-2 are affected by increased coagulopathy, including in the most advanced forms, a fully blown disseminated intravascular coagulation, leading to multi organ failure (MOF).

Post-Morten observations from patients who died because of SARS-CoV-2 infection in Bergamo, Italy and other places (unpublished data) have revealed the presence of diffuse venous, arterial and microcirculatory thrombosis, not only restricted to the lung but also involving the kidneys, heart and gut.

Moreover, elevated D-dimers or low platelet count are independent predictors of morbidity and mortality among SARS-CoV-2 infected patients. [1, 2]

A recent case series demonstrated endothelial cell involvement across vascular beds of different organs in a series of patients with COVID-19. [3]

Inflammation and haemostasis are tightly interrelated pathophysiologic processes that considerably affect each other. In this bidirectional relationship, inflammation leads to activation of the haemostatic system that in turn also considerably influences inflammatory activity. Inflammation shifts the haemostatic activity towards procoagulant state by the ability of proinflammatory mediators to activate coagulation system and to inhibit anticoagulant and fibrinolytic activities. Once the activation of haemostatic system occurs in inflammatory states, amplification of the haemostatic disorder can result in thrombosis and organ damage. In turn, uncontrolled activation of the haemostatic system can also amplify the initial inflammatory response thus causing additional organ injury.

Thrombin plays a central role in mediating clot forming as well as in mediating inflammation. [4]

2. RATIONALE

Therefore, the study aims to assess the value of a direct factor X inhibitor, namely edoxaban as prophylactic measure to mitigate the risk of venous and arterial thrombotic complications and we will explore its effect on markers of inflammation as well as coagulation among SARS-CoV-2 patients at high risk for developing COVID-19 -related morbidity and mortality.

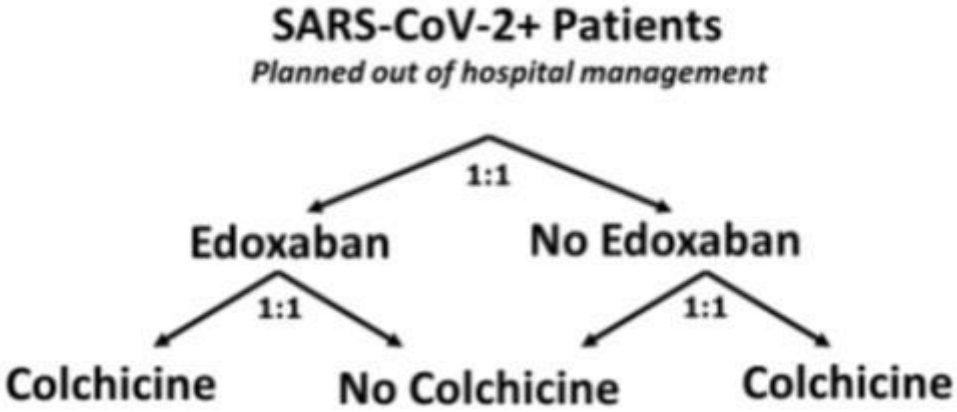
Colchicine has been used since antiquity, with an extensive history of clinical experience in the treatment of many inflammatory diseases (specially in gout). [5] Colchicine has been administered in numerous research protocols with various clinical settings (including stable coronary artery disease, acute myocardial infarction and stroke) showing a favourable safety profile and promising results. [6, 7, 8, 9, 10] In patients with elevated inflammatory markers, colchicine improves endothelial function. [11]

Therefore, Colchicine may exert anti-inflammatory as well as direct anti-viral effects among patients with SARS-CoV-2. Under this rational, multiple studies have been launched testing the value of colchicine in COVID-19 patients. [12] Yet, none will assess whether the use of colchicine accelerates SARS-CoV-2 clearance.

3. STUDY DESIGN

The study will be conducted as a multi-centre, open-label, randomized, 2x2 factorial design clinical trial in SARS-CoV-2 positive patients managed outside the hospital.

Eligible patients will be randomized within seven days from first SARS-CoV-2 positive diagnosis to edoxaban or no edoxaban and to colchicine or no colchicine, as per scheme below:



The randomization procedure is programmed into the eCRF, in random blocks, stratified by clinical site and sex, to first edoxaban versus no treatment and second to colchicine versus no treatment in a 1:1:1:1 ratio.

The study includes 5 visits:
- randomization visit (V1)
- at 7 (+/-3) days post randomization visit (V2), when also phone contact or video call is allowed
- at 14 (+/-3) days post randomization visit (V3), in outpatient clinic
- at 21 (+/-3) days post randomization visit (V4), when also phone contact or video call is allowed
- at 25 (+/-3) days post randomization visit (V5), in outpatient clinic
- Patients with persisting symptoms at 25 (+/-3) day visit will be followed-up on a monthly basis by means of phone calls until at least 7 days after symptom resolution.

4. STUDY OBJECTIVES

The aim of the CONVINCE study is therefore to assess the safety and efficacy of edoxaban and/or colchicine administration in SARS-CoV-2 infected patients who are managed outside the hospital with respect to the occurrence of fatalities, hospitalisation, major vascular thrombotic events or the SARS-CoV-2 clearance rate under RT PCR.

More specifically, the objectives of the study are to test the following hypotheses (independently):

1) The edoxaban regimen is superior to no edoxaban treatment for occurrence of major vascular thrombotic events (MVTE), defined as the composite of asymptomatic proximal deep-vein thrombosis, symptomatic proximal or distal deep-vein thrombosis, symptomatic pulmonary embolism or thrombosis, myocardial infarction, ischemic stroke, non-CNS systemic embolism and all-cause death.
2) The colchicine regimen is superior to no colchicine treatment for the SARS-CoV-2 clearance rates under RT PCR or freedom from death or hospitalisation.

5. ENDPOINTS AND COVARIATES

5.1 Primary endpoints

This study has 2 co-primary endpoints, one each randomization as follows:

Randomization 1 - Edoxaban vs. no active treatment

Major vascular thrombotic events (MVTE) at 25 (+/-3) days defined as a composite of:

- Asymptomatic proximal deep-vein thrombosis
- Symptomatic proximal or distal deep-vein thrombosis
- Symptomatic pulmonary embolism or thrombosis
- Myocardial infarction
- Ischemic stroke
- non-CNS systemic embolism
- Death

Randomization 2 - Colchicine vs no active treatment

The SARS-CoV-2 detection rates at day 14 (+/-3) under RT PCR or freedom from death or hospitalisation

COVID-19 related morbidity and mortality is believed to be the result of a complex interplay between systemic inflammation and hypercoagulation. The factorial study design allows us to test in the same population two hypotheses related to two different treatments, namely colchicine, targeting inflammation and edoxaban, targeting the coagulation pathways. For each experimental treatment, an independent endpoint has been developed and no interaction between the two is expected, making the factorial study design ideal to test two new possible treatment options in the same patient population. The chosen endpoint for edoxaban investigates the potential effect of this treatment on a broad spectrum of thrombotic disorders occurring in the venous or arterial systems whereas the chosen primary endpoint for colchicine captures the possible effect of colchicine on virus clearance or clinical deterioration resulting in hospitalisation or death. The only overlapping endpoint between the two independent primary hypothesis is death.

A recent meta-analysis in COVID-19 patients [13] showed a pooled rate of venous thromboembolism (VTE) of 12.5% in hospitalized patients and 17.2% in intensive care unit patients. Notably all patients received prophylactic anti-coagulation and this study identified full anticoagulation with unfractionated heparin a protective factor for VTE (Pooled odds ratio 0.33 [95% confidence interval 0.14-0.75; P = 0.008], I = 0%).
Another systematic review and meta-analysis [14] reported among hospitalized COVID-19 patients a pooled incidence of vein thromboembolism of 2%, deep vein thrombosis of 17%, pulmonary embolism of 2%, stroke of 1% and myocardial infarction of 6%.
To the best of our knowledge, there is no published information on the incidence of major vascular thrombotic events in symptomatic COVID-19 patients managed in the out of hospital setting. Therefore, our sample size calculation was based on a very conservative estimation (i.e. more than 50% discount) informed by the published data on the rates of major vascular thrombotic events in COVID-19 hospitalized patients receiving prophylactic or full-dosed anticoagulation. Regarding the assumptions taken for the colchicine endpoint, we set the background event rate based on the knowledge that the median period of viral genome detectability for COVID-19 positive patients is 15 days [15] and expected from

colchicine a treatment effect consistent with less than half of what has been previously shown by hydroxychloroquine on viral loads [16]. We did not compute the additional possible colchicine treatment effect on the need for hospitalization to further increase the study power and finally expected a minimal number of fatal events among the non-hospitalized patients recruited in the trial.

5.2 Secondary endpoints

The secondary endpoints of the study are the following:

1) Each component of the co-primary endpoints
2) Need for non-invasive or invasive ventilation
3) Need for oxygen therapy
4) Body Temperature kinetics
5) Need for analgesics including NSAIDs and/or paracetamol
6) Need for hospitalisation and total days in the hospital
7) Any combination of the above endpoints
8) Each component of the primary endpoint as well as pre-specified composite endpoints at the time all SAEs have come to a resolution
9) Global burden of disease defined as duration and severity of symptoms, morbidly and mortality until symptom resolution.
10) Impact of either intervention on coagulation and inflammatory biomarkers including IL-6, CRP, D-dimers, sCD40L, Fibrinogen, Factor X activity and Factor XIa
11) EKG analyses for QT segment measures and for detection of EKC changes associated to myo- pericarditis.
12) HsTroponin levels
13) Bleeding endpoints according to the Bleeding Academic Research Consortium (BARC) 2, 3 or 5 and ISTH major and clinically relevant non-major bleeding

6. ANALYSIS SETS/ POPULATIONS

The following populations will be considered for data analysis:

Full analysis (FAS) population
FAS population consists of all randomized subjects. Subjects are categorized according to the group to which they were assigned by the randomization process.

Per protocol (PP) population
PP population consists of randomized patients who met the following criteria: no violation of inclusion and exclusion criteria and randomized treatment was implemented within 48 hours after randomization.

On-treatment population
On-treatment population consists of randomized patients in whom overall adherence to protocol mandated medications is of more than 80% or less than 120%.

Modified intention-to-treat population (mITT)
mITT population includes randomized subjects in whom at least 1 dose of study medication was administered and have an adequate assessment of MVTE (for the edoxaban versus no edoxaban arm) or an adequate assessment of SARS-CoV-2 infection rate under RT PCR (for colchicine versus no colchicine comparison).

An adequate assessment of MVTE is present if at least one of the following conditions is fulfilled:

- an adequate bilateral lower extremity venous ultrasonography was performed on Day 25 ± 3
- confirmed symptomatic lower extremity DVT up to Day 25 ± 3
- confirmed symptomatic PE up to Day 25 ± 3
- confirmed VTE related death up to Day 25 ± 3
- confirmed myocardial infarction up to Day 25 ± 3
- confirmed ischemic stroke up to Day 25 ± 3
- confirmed non-CNS systemic embolism up to Day 25 ± 3
- death has occurred before Day 25 ± 3

An adequate assessment of SARS-CoV-2 infection rate is present if follow-up RT-PCR is performed at Day 14± 3 or the patient died or was hospitalized.

6.1 Protocol deviations

Investigator will document and explain any Protocol Deviation that occurred during the course of the study. Each deviation will be evaluated to understand if it has the criteria for inclusion in populations mentioned above.

7. STATISTICAL PROCEDURES

Statistical analyses will be carried out using R Statistical Software (Foundation for Statistical Computing, Vienna, Austria).

Treatment groups will be compared, descriptively and graphically in terms of primary and secondary endpoints. Overall, descriptive statistics will be provided in summary tables according to the type of variable summarised:
• for quantitative variables: standard quantitative statistics (N, mean, standard deviation, median, minimum and maximum);
• for qualitative variables: frequency distribution [number of non-missing observations (N) and percentages (%)].

7.1 Baseline characteristics

Demographic data (age, gender, race), SARS CoV-2 infection characteristics (e.g. general/respiratory/gastrointestinal symptoms and signs, clinical evaluation, neurological evaluation, respiratory examination) and all other baseline characteristics (e.g. comorbidities, medical history, medical treatment before the randomization, exams laboratory, arterial blood gases analysis, ECG, bilateral lower extremity venous ultrasonography) recorded at screening will be summarised in descriptive statistics (according to the type of variable) for the randomised subjects, both for the entire sample and separately by groups of treatment.

7.2 Follow-up

7.2.1 Analysis of the primary endpoints

Main analysis of the primary endpoints is performed in the mITT population.

Sensitivity analyses are performed in the ITT, PP and on-treatment populations. Follow-up is censored at the last date of known outcome status or at 28 days, whichever comes first.

Rates of primary endpoints are estimated as the cumulative incidence from the date of randomization to Day 14 ± 3 after randomization for the colchicine comparison and from the date of randomization to Day 25 ± 3 for edoxaban by the Kaplan-Meier methods.

The relative risk ratio (edoxaban/no edoxaban and colchicine versus no colchicine) with respect to the incidence rates of the primary efficacy endpoints will be estimated based on nonstratified estimators using Mantel-Haenszel weights and the corresponding non-stratified asymptotic 2-sided 95% confidence intervals (CI).

7.2.2 Analysis of the secondary endpoints

As for analysis of primary endpoints, analysis of the secondary endpoints is performed in the mITT population. Sensitivity analyses are performed in the ITT, PP and on-treatment populations.

According to the type of the endpoint, the more appropriate analyses will be performed: Kaplan-Meier methods will be used for qualitative endpoints; anova for repeated measures will be applied to quantitative variables.
For the analysis of the global burden of disease, which will be expressed in arbitrary units (AU) and shown on daily basis and cumulatively, we prespecify to weigh mortality, morbidity and symptoms.
We will assign 1000 AU to death, 700 AU to stroke, and 500 AU to symptomatic pulmonary embolism or thrombosis, or to myocardial infarction or non-CNS systemic embolism, 300 AU to symptomatic proximal or distal deep-vein thrombosis and 200 AU to asymptomatic proximal deep-vein thrombosis. For each day of follow-up from randomization, symptoms will be graded according to none (0 AU), mild (1 AU), moderate (2 AU) or severe (3 AU). For each day in the hospital, an AU of 5 will be assigned incrementally to event and symptom weighing. The severity of symptoms will be self-reported by each included patient. Fever will be graded daily in terms of 5 AU if below 38° or need for up to 1.5 gr paracetamol or NSDAID equivalent, 10 AU in between 38° but <39°C or need for > 1.5 but < or equal 3 grams paracetamol or NSDAID equivalent and 20 AU if > or equal 39°C or need for > 3 grams paracetamol or NSDAID equivalent. Need for oxygen or Sp02 below 90% will generate 10 AU. Non-invasive ventilation will be graded 50 AU daily, whereas invasive ventilation 100 AU daily.

7.2.3 Analysis of safety data (adverse events)

All adverse events will be coded to a Preferred Term (PT) and Primary System Organ Class (SOC), using the MedDRA dictionary using the current version. AEs will be tabulated as PT, SOC and frequency for mITT population, both for the entire sample and separately by groups of treatment. Only descriptive analysis is planned.

7.2.4 Subgroup analyses

The following subgroups are pre-specified:

- Age ≥65 years vs. age <65 years
- Female vs. male gender
- High vs. non-high risk
- Creatinine clearance equal or greater than 50 ml/min or < 50 ml/min or fulfilment of other dose reduction criteria for edoxaban
- Duration of COVID-19 symptoms to recruitment stratified by median value
- According to presence or absence of each high-risk criteria
- Allocation to edoxaban versus no edoxaban for colchicine randomisation and to colchicine versus no colchicine for edoxaban randomisation.

High risk criteria include the following conditions:

1) Active malignancy
2) Prior myocardial infarction
3) History of heart failure
4) Diabetes mellitus
5) D-dimers above the upper limit of normal
6) Active smoking
7) Chronic obstructive pulmonary disease or other chronic lung diseases (e.g. pulmonary fibrosis)
8) Prior thrombo-embolic disorder
9) Other conditions which at discretion of the investigator put the patient at higher risk

for COVID-19 related complications such as but not limited to severity of symptoms
or hs-troponin elevation. This information will be recorded in the eCRF at the time of inclusion.

8. REFERENCES

[1] Tang N, Li D, Wang X, Sun Z. Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. J Thromb Haemost2020;18(4):844-847.

[2] Zhou F, Fan G, Liu Z, Cao B. SARS-CoV-2 shedding and infectivity - Authors' reply. Lancet 2020.

[3] Varga Z, Flammer AJ, Steiger P, Haberecker M, Andermatt R, Zinkernagel AS, Mehra MR, Schuepbach RA, Ruschitzka F, Moch H. Endothelial cell infection and endotheliitis inCOVID-19. Lancet 2020.

[4] Posma JJ, Posthuma JJ, Spronk HM. Coagulation and non-coagulation effects of thrombin. J Thromb Haemost 2016;14(10):1908-1916.

[5] Niel E, Scherrmann JM. Colchicine today. Joint Bone Spine 2006;73(6):672-8.

[6] Hemkens LG, Ewald H, Gloy VL, Arpagaus A, Olu KK, Nidorf M, Glinz D, Nordmann AJ,Briel M. Cardiovascular effects and safety of long-term colchicine treatment: Cochrane review and meta-analysis. Heart 2016;102(8):590-6.

[7] Nidorf SM, Fiolet ATL, Eikelboom JW, Schut A, Opstal TSJ, Bax WA, Budgeon CA, Tijssen JGP, Mosterd A, Cornel JH, Thompson PL, LoDoCo I. The effect of low-dose colchicine in patients with stable coronary artery disease: The LoDoCo2 trial rationale, design, and baseline characteristics. Am Heart J 2019;218:46-56.

[8] Tucker B, Kurup R, Barraclough J, Henriquez R, Cartland S, Arnott C, Misra A, Martinez G, Kavurma M, Patel S. Colchicine as a Novel Therapy for Suppressing Chemokine Production in Patients With an Acute Coronary Syndrome: A Pilot Study. Clin Ther 2019;41(10):2172-2181.

[9] Khandkar C, Vaidya K, Patel S. Colchicine for Stroke Prevention: A Systematic Review and Meta-analysis. Clin Ther 2019;41(3):582-590 e3.

[10] Tardif JC, Kouz S, Waters DD, Bertrand OF, Diaz R, Maggioni AP, Pinto FJ, Ibrahim R, Gamra H, Kiwan GS, Berry C, Lopez-Sendon J, Ostadal P, Koenig W, Angoulvant D, Gregoire JC, Lavoie MA, Dube MP, Rhainds D, Provencher M, Blondeau L, Orfanos A, L'Allier PL, Guertin MC, Roubille F. Efficacy and Safety of Low-Dose Colchicine after Myocardial Infarction. N Engl J Med 2019;381(26):2497-2505.

[11] Kajikawa M, Higashi Y, Tomiyama H, Maruhashi T, Kurisu S, Kihara Y, Mutoh A, Ueda SI. Effect of short-term colchicine treatment on endothelial function in patients with coronary artery disease. Int J Cardiol 2019;281:35-39.

[12] Deftereos SG, Siasos G, Giannopoulos G, Vrachatis DA, Angelidis C, Giotaki SG, Gargalianos P, Giamarellou H, Gogos C, Daikos G, Lazanas M, Lagiou P, Saroglou G, Sipsas N, Tsiodras S, Chatzigeorgiou D, Moussas N, Kotanidou A, Koulouris N, Oikonomou E, Kaoukis A, Kossyvakis C, Raisakis K, Fountoulaki K, Comis M, Tsiachris D, Sarri E, Theodorakis A, Martinez-Dolz L, Sanz-Sanchez J, Reimers B, Stefanini GG, Cleman M, Filippou D, Olympios CD, Pyrgakis VN, Goudevenos J, Hahalis G, Kolettis TM, Iliodromitis E, Tousoulis D, Stefanadis C. The Greek study in the effects of colchicine in COvid-19 complications prevention (GRECCO-19 study): Rationale and study design. Hellenic J Cardiol 2020.

[13] Sridharan GK, Vegunta R, Rokkam VRP, Meyyur Aravamudan V, Vegunta R, Khan SR, Ponnada S, Boregowda U, Prudhvi K, Chamarthi G, Mohan BP. Venous Thromboembolism in Hospitalized COVID-19 Patients. Am J Ther. 2020 Nov/Dec;27(6):e599-e610. doi: 10.1097/MJT.0000000000001295. PMID: 33156016.

[14] McBane RD 2nd, Torres Roldan VD, Niven AS, Pruthi RK, Franco PM, Linderbaum JA, Casanegra AI, Oyen LJ, Houghton DE, Marshall AL, Ou NN, Siegel JL, Wysokinski WE, Padrnos LJ, Rivera CE, Flo GL, Shamoun FE, Silvers SM, Nayfeh T, Urtecho M, Shah S, Benkhadra R, Saadi SM, Firwana M, Jawaid T, Amin M, Prokop LJ, Murad MH. Anticoagulation in COVID-19: A Systematic Review, Meta-analysis, and Rapid Guidance From Mayo Clinic. Mayo Clin Proc. 2020 Nov;95(11):2467-2486. doi: 10.1016/j.mayocp.2020.08.030. Epub 2020 Aug 31. PMID: 33153635.

[15] Cimrman Š, Macková L, Král V, Bartoš H, Stiborová I, Dlouhý P. The duration of SARS-CoV-2 shedding in patients recovering from COVID-19. Epidemiol Mikrobiol Imunol. 2020 Summer;69(3):148-151. English. PMID: 33086857.

[16] Gautret P, Lagier JC, Parola P, Hoang VT, Meddeb L, Mailhe M, Doudier B, Courjon J, Giordanengo V, Vieira VE, Tissot Dupont H, Honoré S, Colson P, Chabrière E, La Scola B, Rolain JM, Brouqui P, Raoult D. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. Int J Antimicrob Agents. 2020 Jul;56(1):105949. doi: 10.1016/j.ijantimicag.2020.105949. Epub 2020 Mar 20. PMID: 32205204; PMCID: PMC7102549.